In VitroToxicity Evaluation of Nanomaterials: Importance of Materials Characterization

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WPAFB 08-5128

Report Documentation Page

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
28 MAR 2011		**00-00-2011 to 00-00-2011**

4. TITLE AND SUBTITLE	5a. CONTRACT NUMBER
In VitroToxicity Evaluation of Nanomaterials: Importance of Materials Characterization	5b. GRANT NUMBER
	5c. PROGRAM ELEMENT NUMBER
6. AUTHOR(S)	5d. PROJECT NUMBER
	5e. TASK NUMBER
	5f. WORK UNIT NUMBER
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) **Air Force Research Laboratory,Human Effectiveness Directorate,Wright-Patterson AFB,OH,45433**	8. PERFORMING ORGANIZATION REPORT NUMBER
9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)	10. SPONSOR/MONITOR'S ACRONYM(S)
	11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION/AVAILABILITY STATEMENT
Approved for public release; distribution unlimited

13. SUPPLEMENTARY NOTES
Presented at the 2011 DoD Environmental Monitoring & Data Quality Workshop (EMDQ 2011), 28 Mar ? 1 Apr, Arlington, VA.

14. ABSTRACT

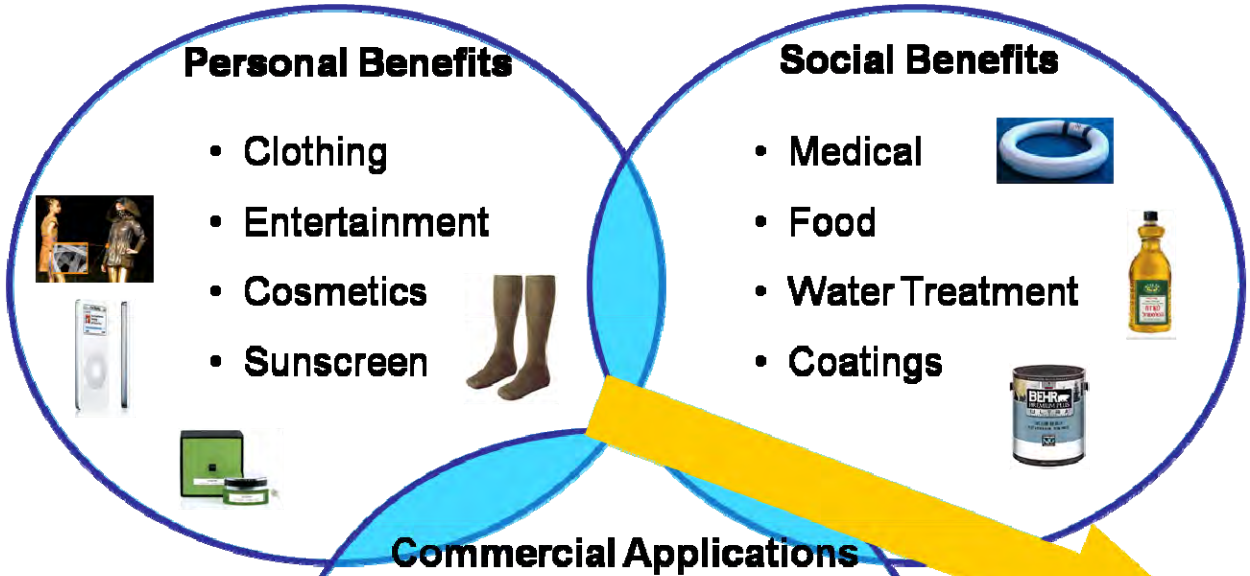
15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT **unclassified**	b. ABSTRACT **unclassified**	c. THIS PAGE **unclassified**	**Same as Report (SAR)**	**57**	

Outline of Presentation

- **Overview**
- **Biological Interaction of NM**
- **Characterization-Technical Challenges**
- **Toxicity Response of NM**
  - Size
  - Surface Coating
  - Charge
  - Shape or Structure
- **Summary and Conclusion**

Applications for Nanotechnology



DoD Applications

•Biosensors
•Antimicrobial Agents
•Munitions
•Propellents
•Coatings
•Smart suites

Applications for Nanotechnology:Silver

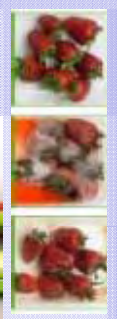
Nanosilver in Footwear

http://www.nanosilverproducts.com/

http://www.latimes.com/features/health/la-he-nanosilver4-2008aug04,0,3206871.story

Uses of Ag NPs

Antibacterial Nanosilver Infused in Storage Containers



Day 1

Day 8

Day 8 w/ Fresher Longer

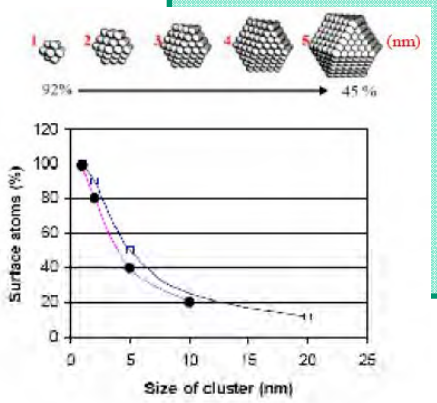
Nanosilver and Antimicrobial Personal Care

Nanosilver Coated Surfaces of Medical Devices to Reduce Hospital Related Infections

Unique Properties of Nanoparticles

Unique Properties

- Optical (metal & Semiconductors
- Magnetic (metal)
- Electronic & Thermal (CN)
- Catalytic activity (high surface area)



Journal of Physics Science and Idea 4 (2004) No. 1 & 2

Challenges

- Controlled synthesis
  - Nano size (1-100 nm)
  - Large surface area relative to mass
  - Surface chemistry and dissolution
  - Surface reactivity/bioaffinities
  - Surface energy
  - Shape/Dimensional Character
- Toxicity and biocompatibility

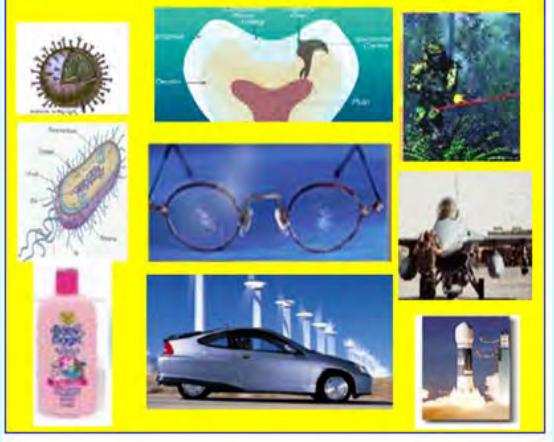
Do We Know Potential Risk?

What are Potential Routes of Exposure?

Potential Routes of Exposure

Application

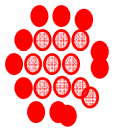
- Nanoenergetic Materials
- Propellants & Munition
- Ultralight Soldier Clothing
- Chemical & Biological Defense
- Diagnostic and screening
- Drug delivery devices
- Optical Coating
- Consumer Products
- Antimicrobials



Bionanotechnology-Tissue engineering, Cognition

Exposure ?

Nanoparticles - Work Environment ??



Transport:Food chain & Bioavailability

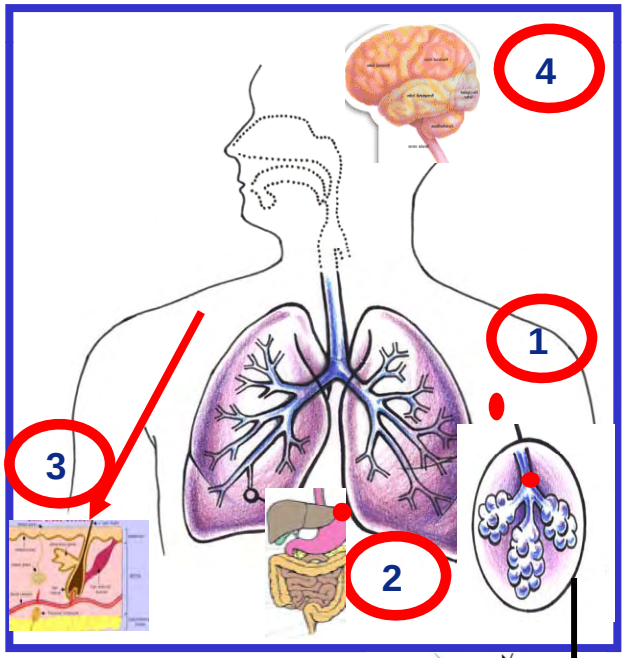
- NP release
- Distribution (water, soil, air)
- Bio-accumulation/persistence

Implication

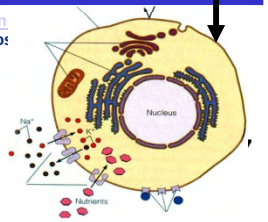
Human health effects of nanomaterials?

"Sufficient data is lacking"

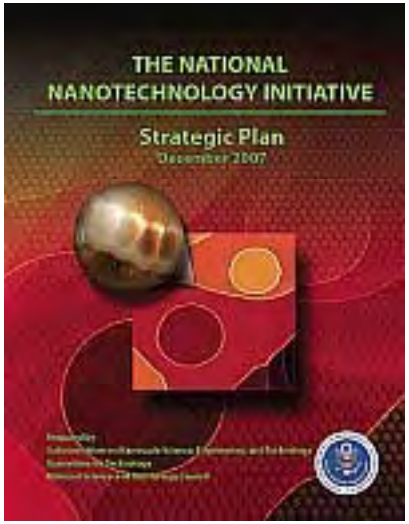
Risk Assessment ?

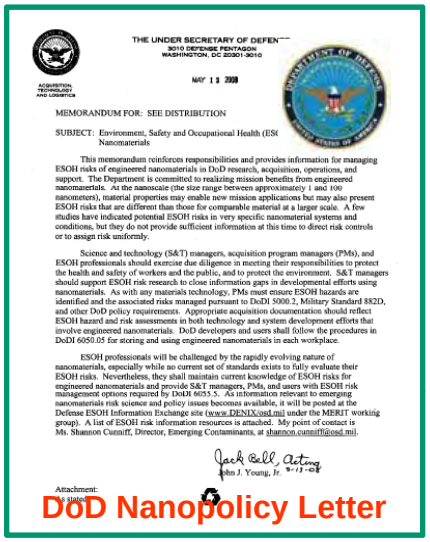


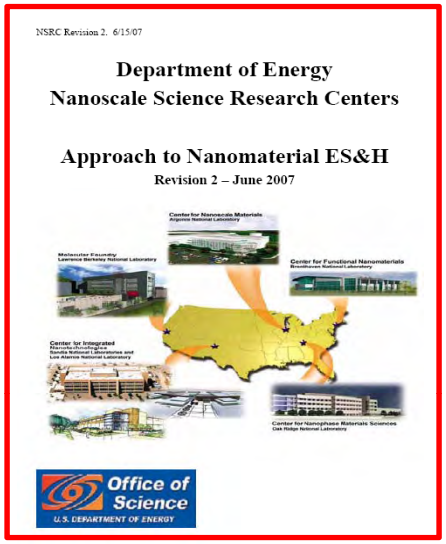
http://www.enchantedlearn
subjects/anatomy/skin/cros

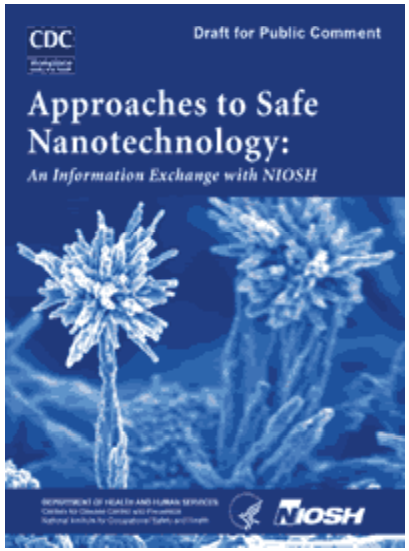


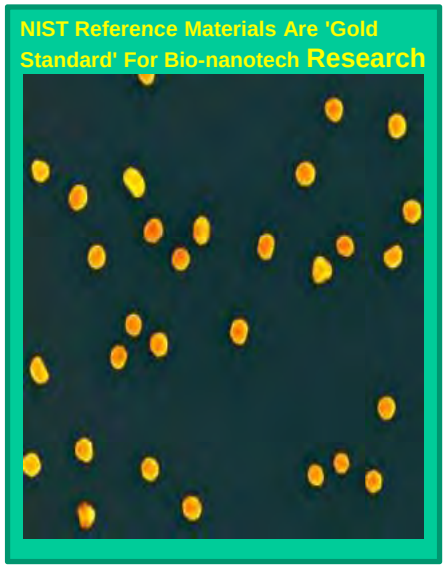
Government Departments and Agencies Participating in Nanotoxicity







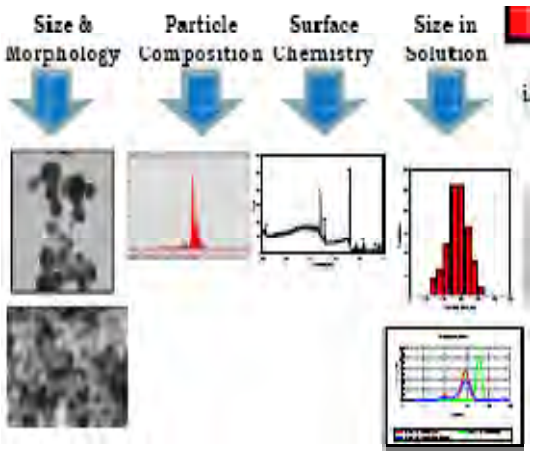




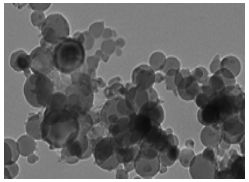
Basic Understanding of Biological Interaction of NM

Biological Interaction of Nanomaterials: Process Overview

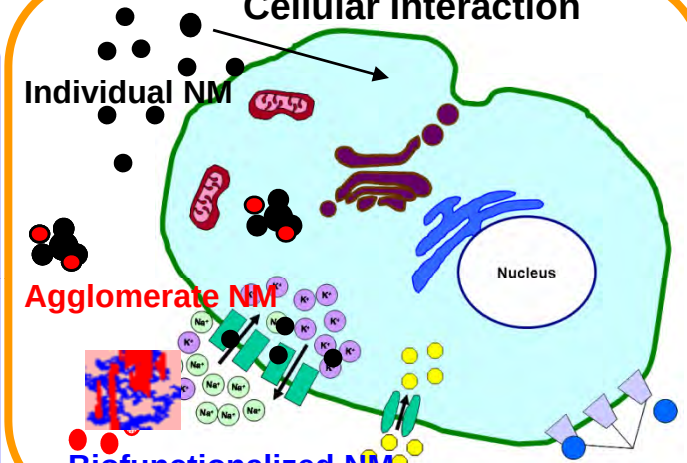
Characterization



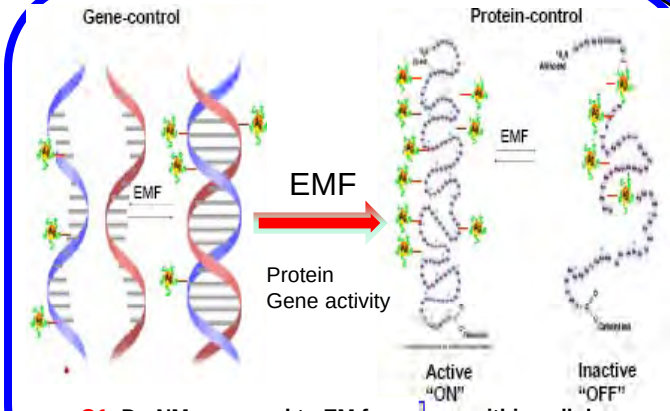
Sample	Average Nanoparticle Size after Dispersion in Lung Surfactant	
	TEM (nm)	DLSZ-Ave (d.nm) ± PdI
Al_2O_3 40nm	48.08 ± 21.01	878 ± 0.495
Al 50nm	32.71 ± 28.28	805 ± 0.497
Al-OA 50nm	51.09 ± 22.48	2430 ± 1.00



Bio-Nano-Interaction



Beneficial Effects



Q1: Do NMs respond to EM frequency within cellular environment?

Q2: Can we control and manipulate cellular env

Toxicological Effects

In vitro Toxicity

Q1: Uptake?
Q2: Proteins or nucleic acids
Q3: Internal organelles?
Q4: Overall effect to cell function ?
Q5: EM frequency Effect ?

In vivo Toxicity

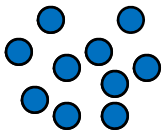
Q1: Exposure?
Q2: Dose
Q3: Acute?
Q4: Chronic ?
Q5: EM frequency respond ?

Nanoparticle Characterization

Predictive Modeling

Post Exposure Characterization of Nanoparticles

Physical Factors:



Disperse?

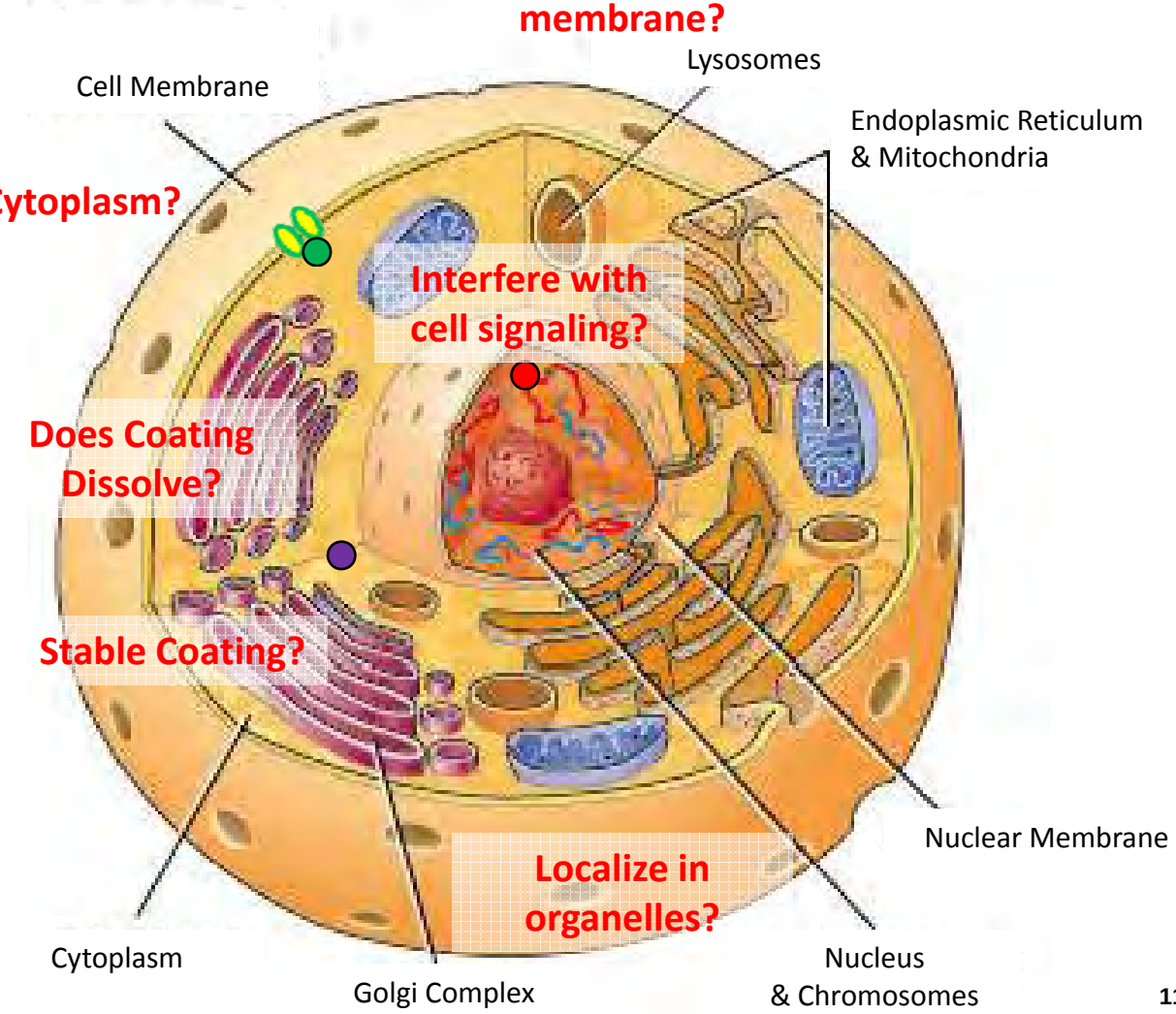
Agglomerate?

Shape?

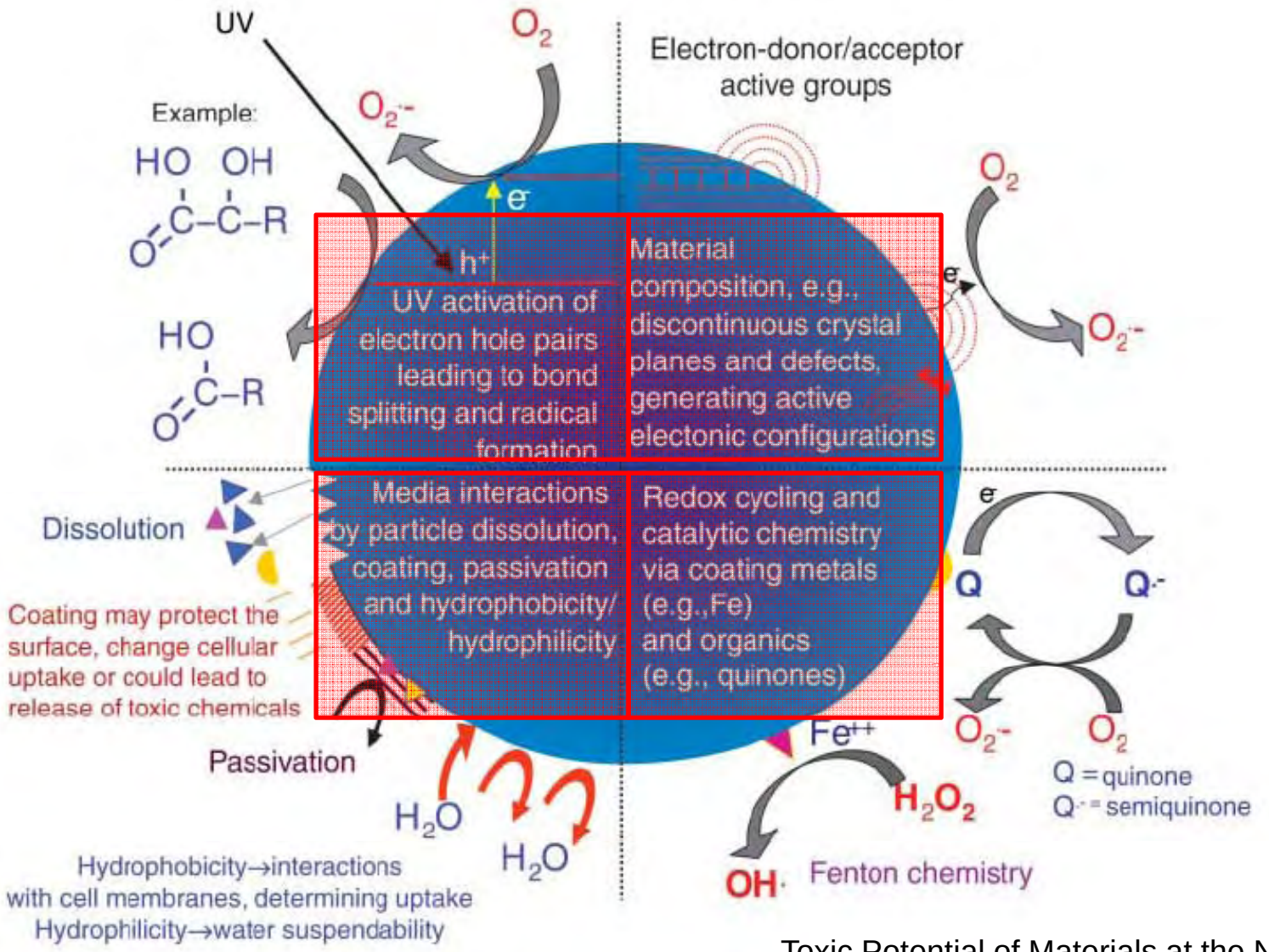
Coating?

Composition?

Affects to Cell:

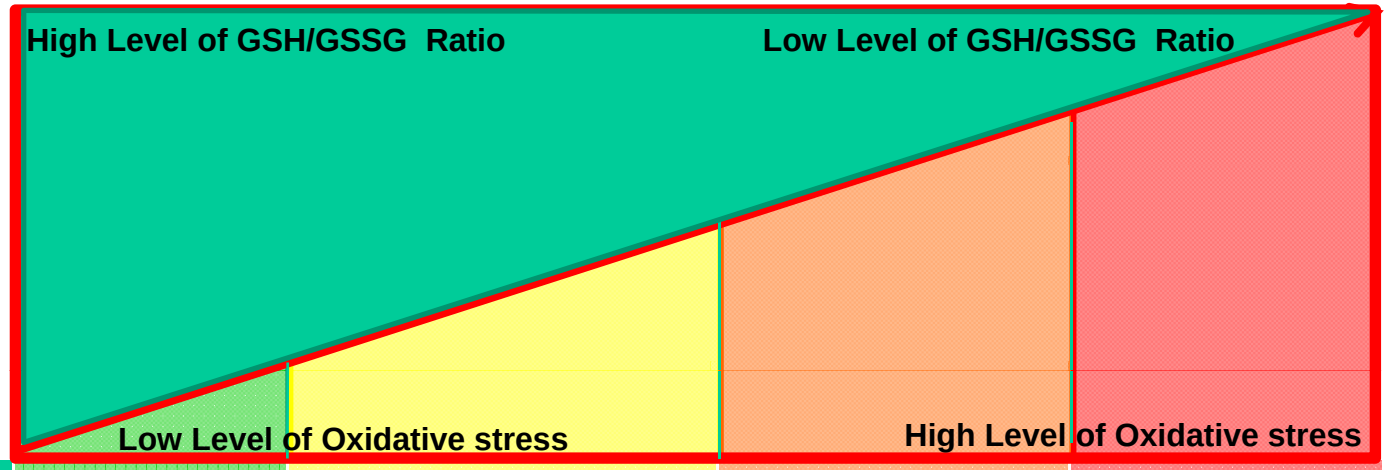


Possible mechanisms by which NM interact with biological tissue



Toxic Potential of Materials at the Nano level.
Andre Nel et al (2006) Feb 3, 311; 622; Science

The Hierarchical Oxidative Stress Model

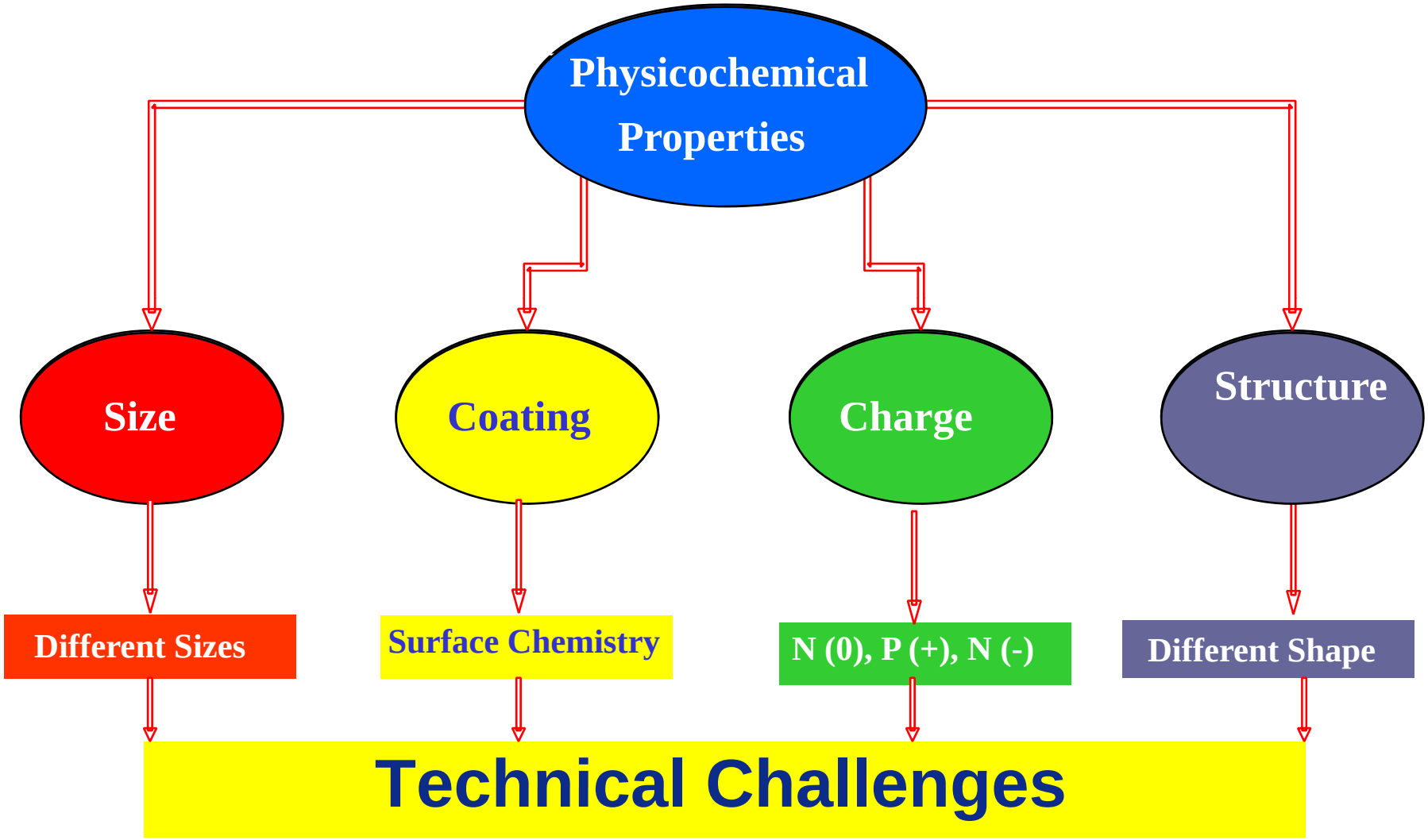


Response Pathways	Normal	I Anti Oxidant Defense	II Pro-inflammatory response	III Cytotoxicity
Signaling	-	Nrf-2	MAP Kinase NF-kB cascade	MMP Electron TS
Gene Exp	-	Anti-oxidant response element	AP-1 NF-kB	-
Outcome	-	Antioxidant enzymes or Phase II enzymes	Cytokines & Chemokines	Necrosis & Apoptosis

Toxic Potential of Materials at the Nano level.
Andre Nel et al (2006) Feb 3, 311; 622; Science

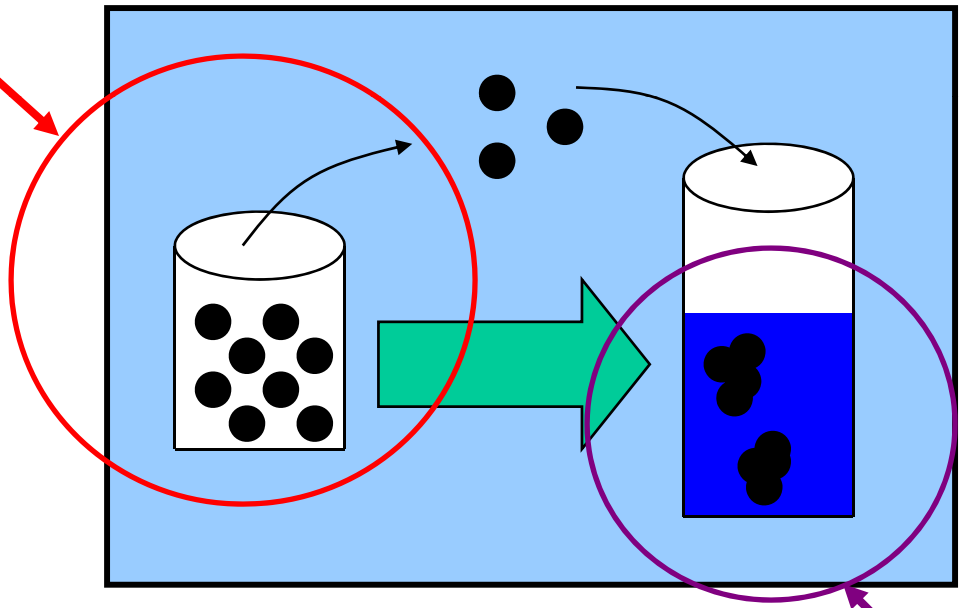
Characterization & Challenges

Toxicity Based on Physicochemical Properties



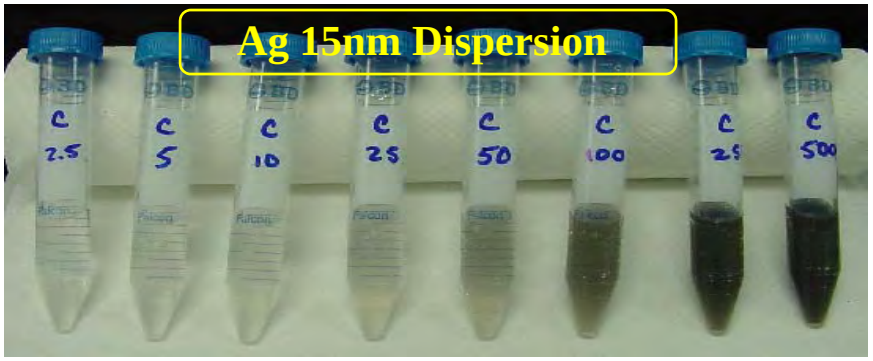
Nanoparticle Size vs. Toxicity

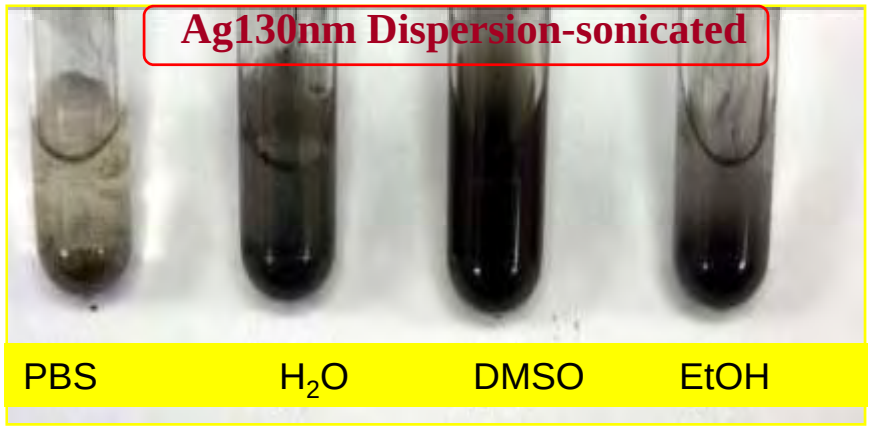
Is the primary size related to the toxicity?

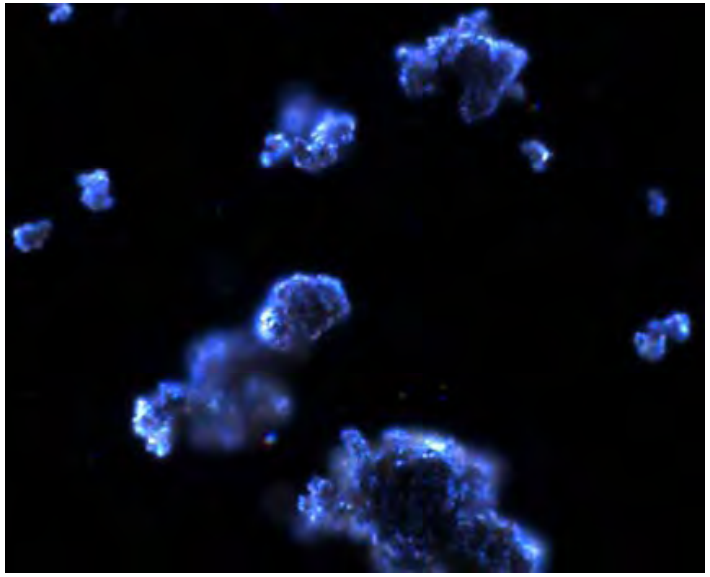


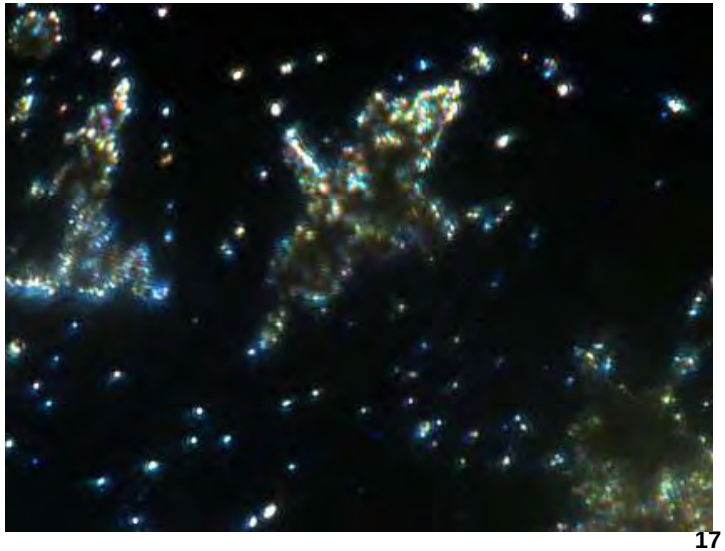
Or is the agglomerated size related to the toxicity?

Turbidity & Dispersion Issues



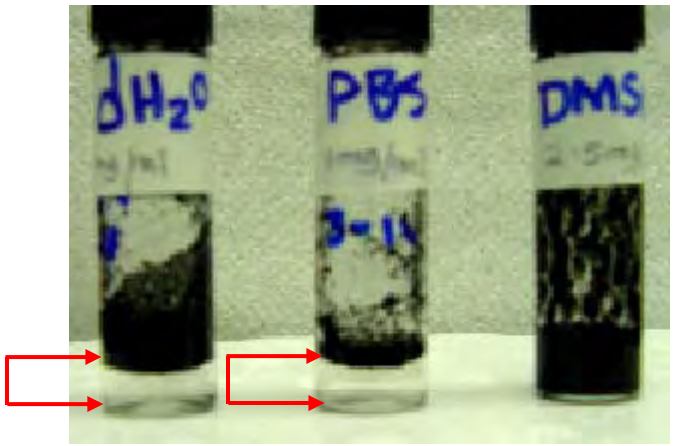




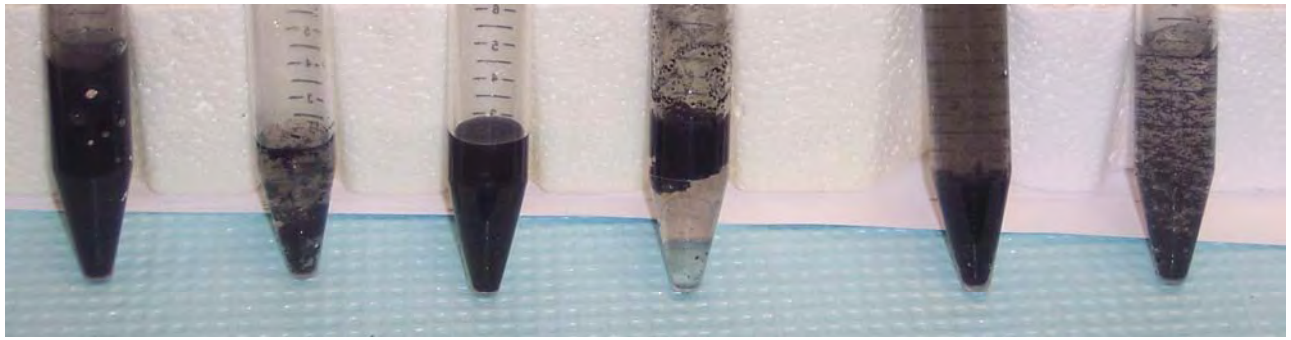


Non-homogeneous Dispersion & Agglomeration

dH2O PBS DMSO

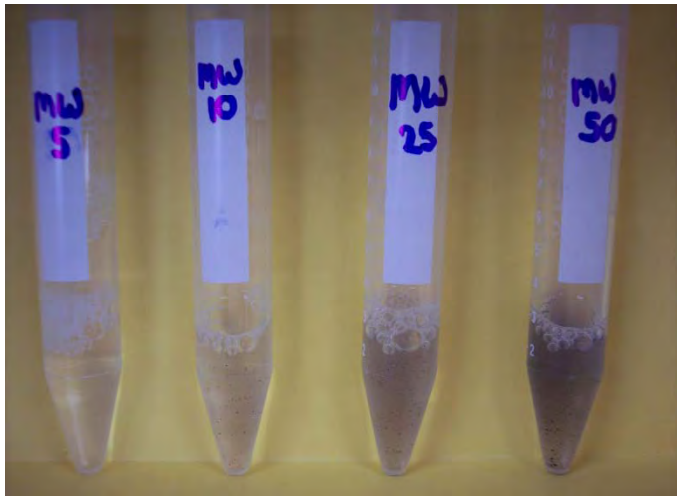


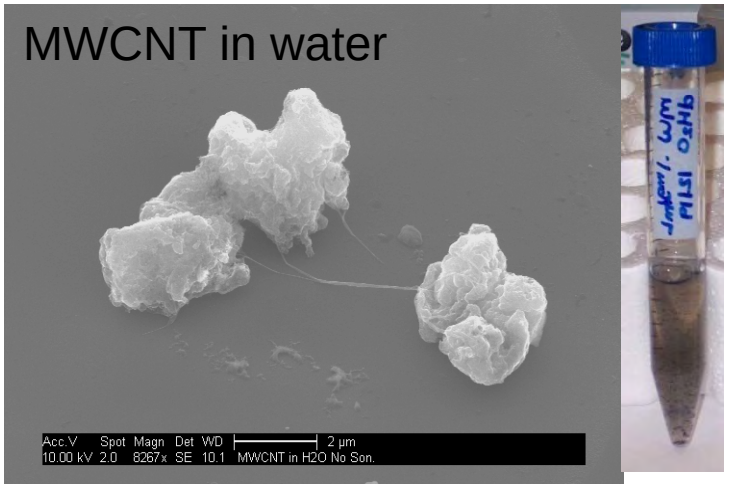
Single Walled Carbon Nanotubes

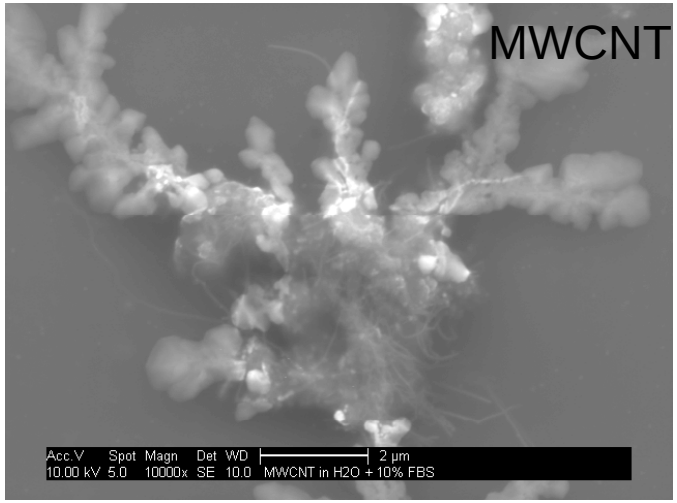


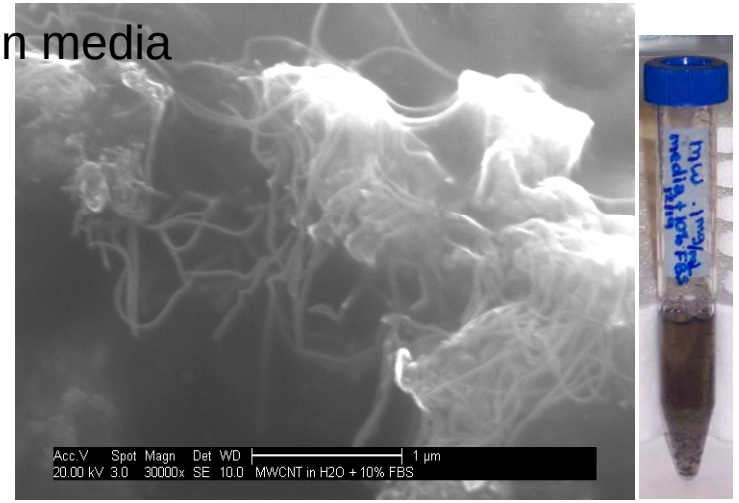
Carbon Nanofibers

MWCNT Agglomerate Structure in Solution

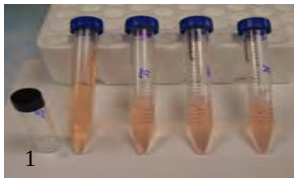




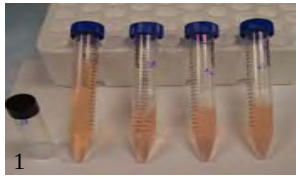




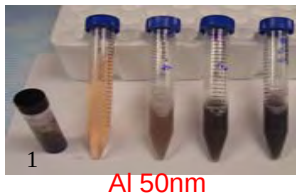
Agglomeration Issues



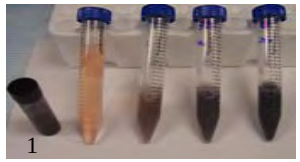
Al2O3 30nm



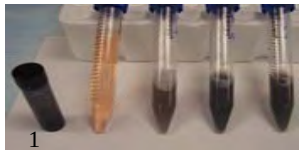
Al2O3 40nm



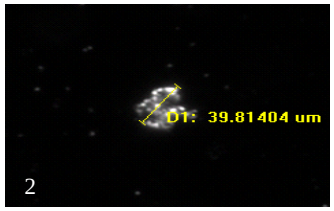
Al 50nm

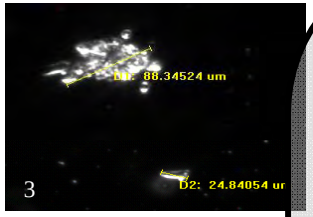


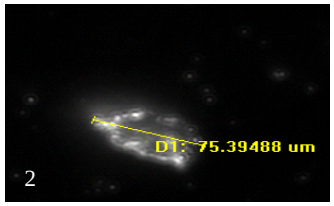
Al 80nm

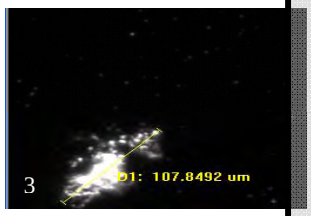


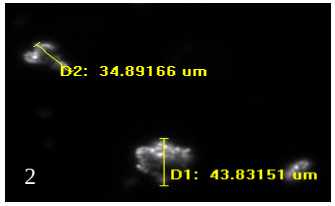
Al 120nm

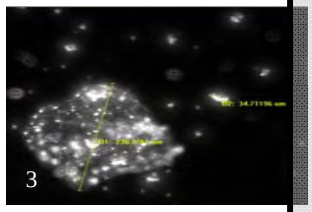


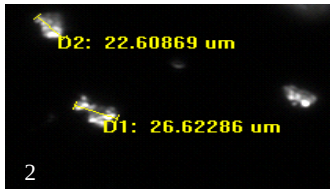


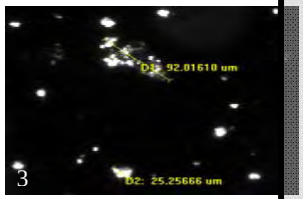


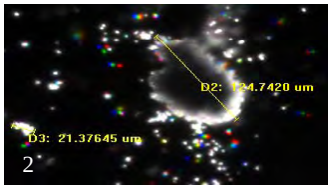


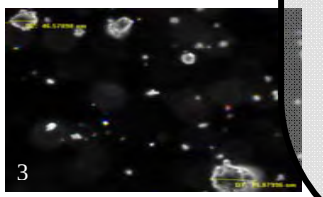












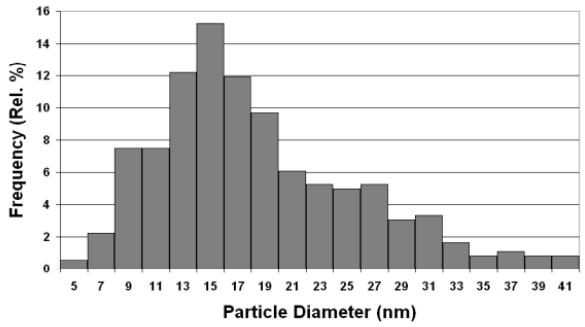
Dynamic Light Scattering

	DLS	
Particle	Diameter (nm)	PDI
Al_2O_3 30nm		
Water	210	0.125
Media	1430	0.373
Media w/ 20% Serum	223	0.23
Al_2O_3 40nm		
Water	237	0.145
Media	1050	0.232
Media w/ 20% Serum	251	0.252
Al 50nm		
Water	253	0.224
Media	1170	0.247
Media w/ 20% Serum	395	0.393
Al 80nm		
Water	378	0.422
Media	1390	0.268
Media w/ 20% Serum	355	0.398
Al 120nm		
Water	342	0.341
Media	1610	0.25
Media w/ 20% Serum	535	0.821

DLS trend of particles highly agglomerating in media without serum, and then decreasing agglomeration with presence of serum.

TEM Measured Size Distributions

A



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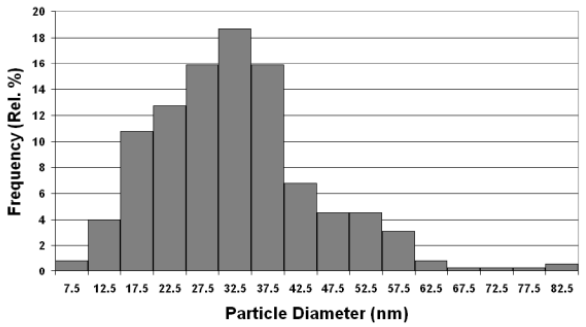

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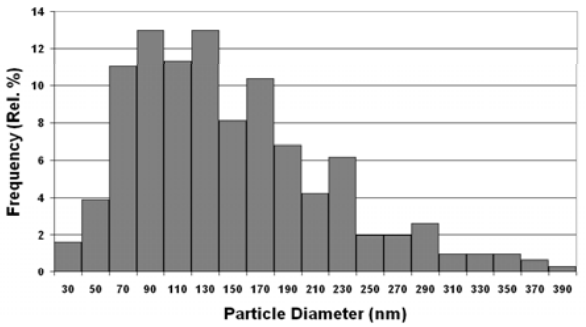

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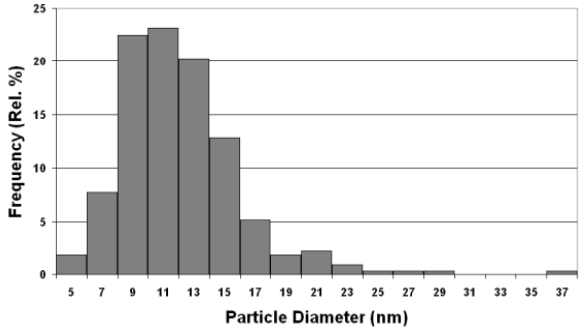
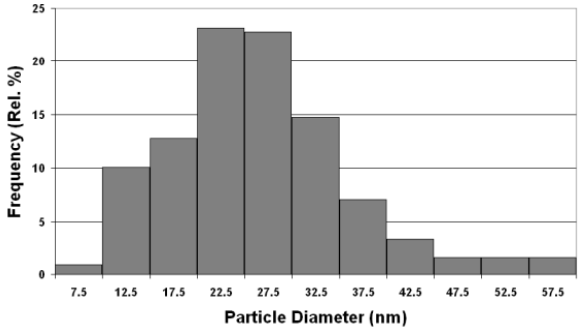


E



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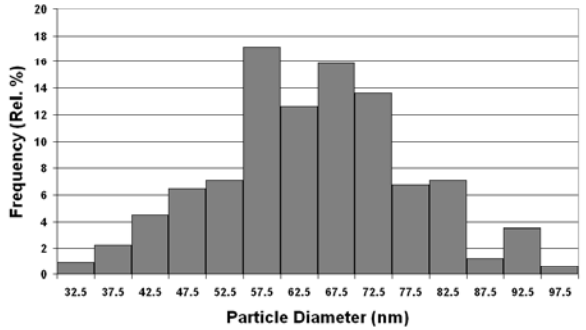

Constant Mass of Sample: 10µg

Sample	Estimate of Total Number of Nanoparticles	Estimate of Surface Area Per Particle (nm^2)	Estimate of Volume Per Particle (nm^3)	Surface Area to Volume Ratio
Ag 15nm	7.8E+11	469	1226	0.382
Ag 25nm	1.6E+11	1267	5840	0.217
Ag 80nm	3.0E+09	16818	321920	0.052
PS-Ag 10nm	1.8E+12	283	519	0.546
PS-Ag 25nm	2.5E+11	997	3824	0.261
PS-Ag 80nm	9.2E+09	10143	104010	0.098

Constant Number of Particles: $1.00x10^{12}$

Sample	Estimate of Mass (mg/mL)	Estimate of Total Surface Area (nm^2)	Estimate of Total Volume (nm^3)
Ag 15nm	0.05	1.2E+15	5.0E+15
Ag 25nm	0.28	3.8E+15	2.7E+16
Ag 80nm	32.23	8.5E+16	3.1E+18
PS-Ag 10nm	0.01	5.1E+14	1.3E+15
PS-Ag 25nm	0.15	2.5E+15	1.4E+16
PS-Ag 80nm	1.65	1.4E+16	1.6E+17

Summary: Technical Challenges

- Variation in size distribution
- Non-homogeneous dispersion
  - Maintaining homogenous dispersion
  - Stability of solution
- Agglomeration after gentle mixing
  - Proper mixing protocols need to be developed
  - Effect of carbon coating
- Increasing turbidity
  - Turbidity may have impact on cells

Collaboration between materials scientists and toxicologists is key to establish safety risk of nanotechnology

Editorial Highlight in Toxicological Sciences
IMPACT

TOXICOLOGICAL HIGHLIGHT

How Meaningful are the Results of Nanotoxicity Studies in the Absence of Adequate Material Characterization?

Editor

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Received November 6, 2007; accepted November 6, 2007

In their publication in this issue, Murdock *et al.* (2007) have focused on the importance of developing adequate physico-chemical characterization of nanomaterials prior to undertaking experiments for *in vitro* toxicity assessments. These authors have correctly suggested that for *in vitro* toxicity studies, particle size, size distribution, particle morphology, particle composition, surface area, surface chemistry, and particle reactivity in solution are important factors which need to be accurately characterized as prerequisites for implementing nanoparticle toxicity studies. This point cannot be overstated,

Therefore, in the Murdock *et al.* study, these investigators have focused on characterizing a wide range of nanomaterials including metals, metal oxides, and carbon-based structures using dynamic light scattering (DLS) concomitant with transmission electron microscopy, for particles dispersed under wet conditions in cell culture media, with and without serum. Some basic cell viability and morphology studies were correlated with DLS particle size characteristic experiments to assess toxicity from observed agglomeration alterations under the various experimental conditions.

Murdock and coworkers concluded that many metals and metal oxide nanomaterials tend to agglomerate in solution. Moreover, other variables, such as the addition of serum in the culture media, can affect toxicity measurements, likely due to influences affecting agglomeration and/or surface chemistry of nanoparticles. These factors represent important considerations that have not been previously recognized.

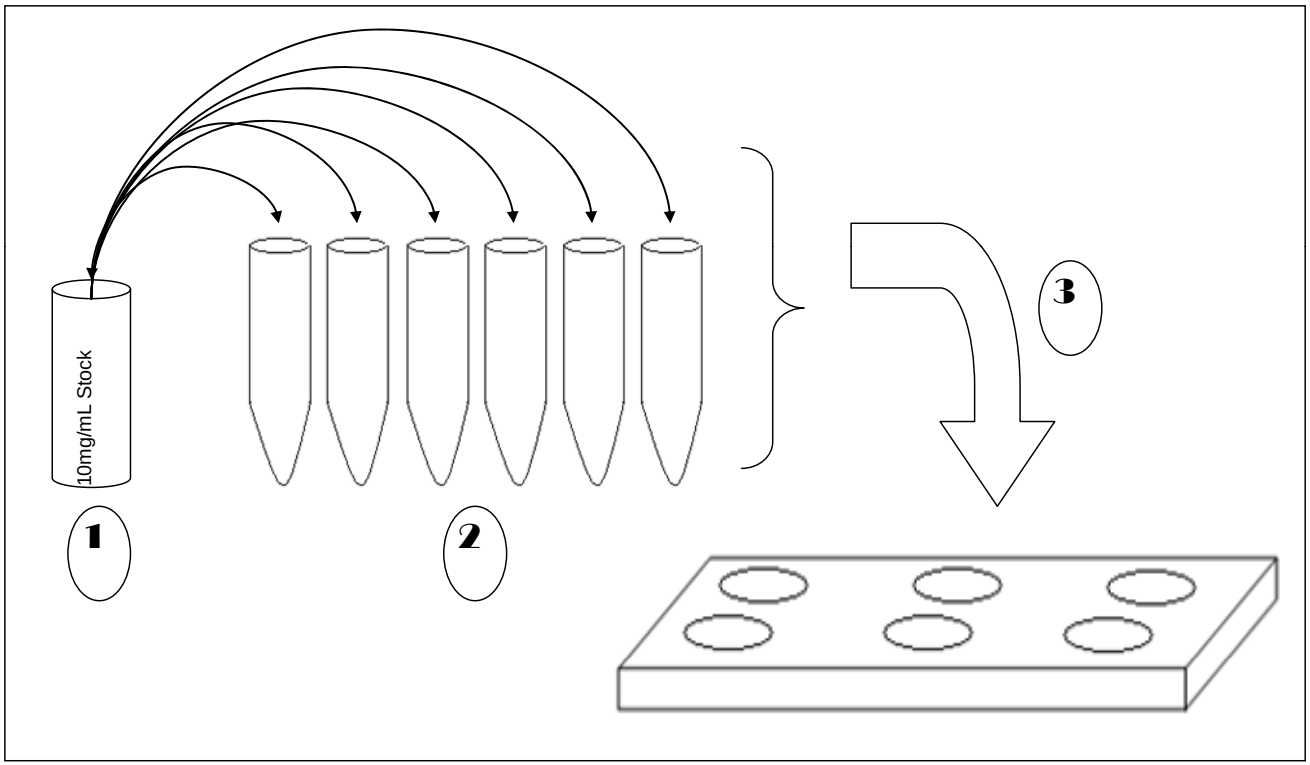
Perhaps the most significant impact of the Murdock *et al.* publication is to raise the issue of the importance of adequately characterizing the nanomaterial preparation prior to the initiation of toxicological experimentation.

Murdock RC, Braydich-Stolle L, Schrand AM, Schlager JJ and Hussain SM (2007) Characterization of nanomaterial dispersion in solution prior to in vitro exposure using dynamic light scattering technique. Toxicol Sci 101:239-253.

Nanotoxicity Studies Experimental Design

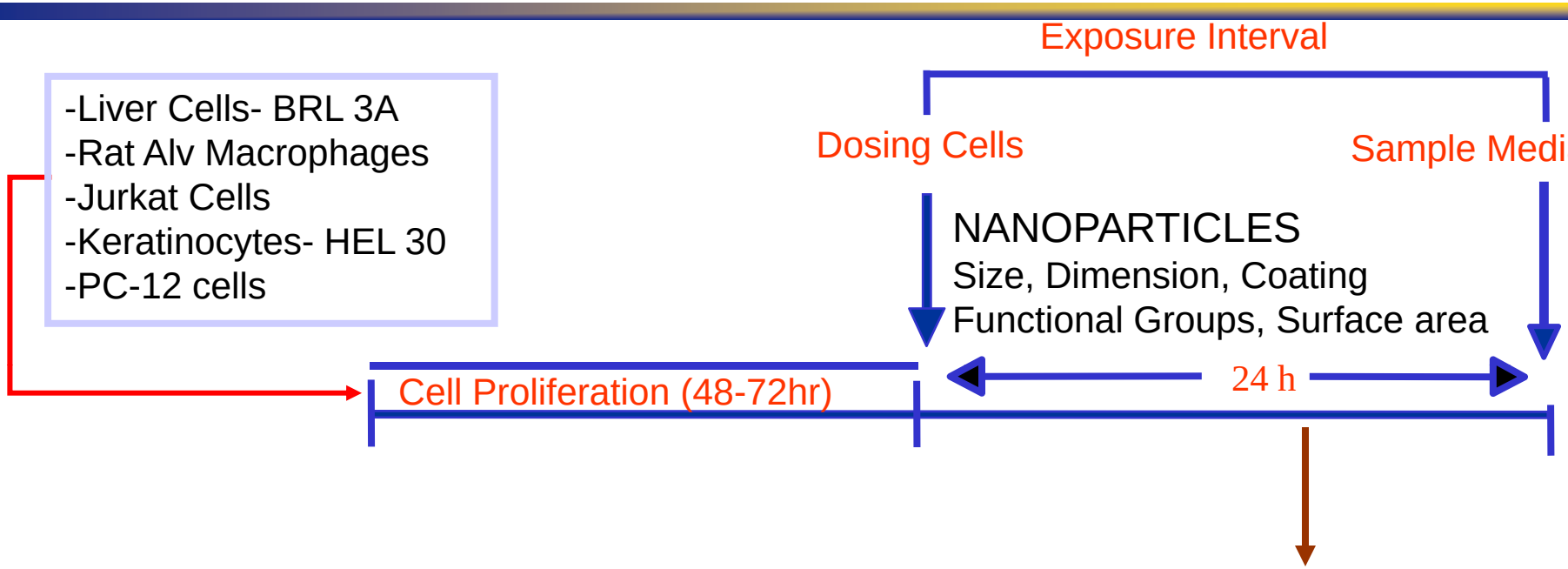
Schematic Representation of Dosing Cells

Nanoparticles



In Vitro Cell Culture

Experimental Design



Toxicity Endpoints

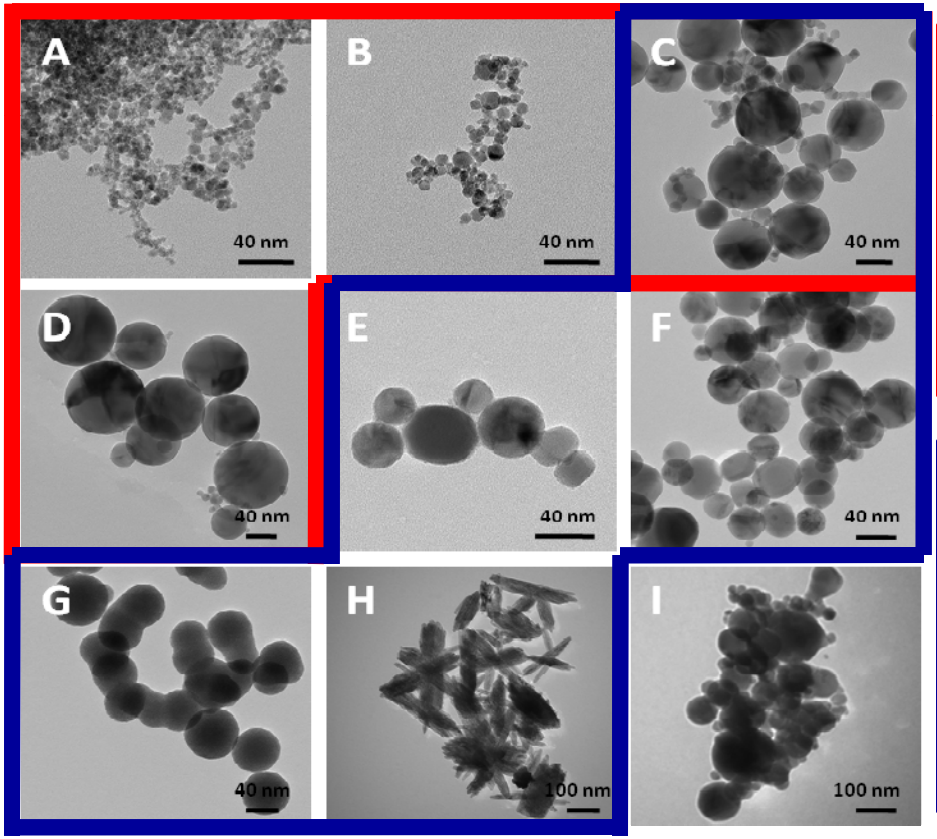
- Membrane Leakage- LDH
- Mitochondrial Function- MTT
- Reactive Oxygen Species- ROS
- Mitochondrial Mem Pot- MMP
- Cytokines- TNF-α, IL-6, MIP-2
- Gene Expression
- Protein Expression

Nanoparticles Size vs. Toxicity

Size vs. Crystal Structure in TiO_2 Nanotoxicity

Study Design

The bioeffects of TiO_2 were studied in mouse keratinocytes using the following nanoparticles:



Size Dependent Study with 100% Anatase TiO_2

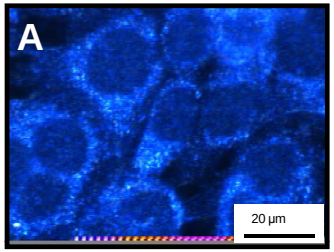
A: 6.3 nm
B: 10 nm
C: 50 nm
D: 100 nm

Crystal Structure Study with TiO_2

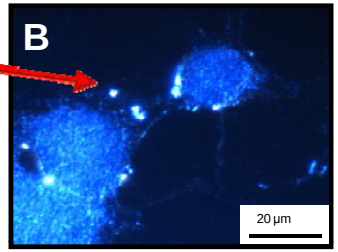
C: 100% Anatase 50 nm
E: 40% Anatase 39 nm
F: 61% Anatase 39 nm
G: Amorphouse 40 nm
H: 100% Rutile 51 nm

Size vs. Crystal Structure in TiO_2 Nanotoxicity

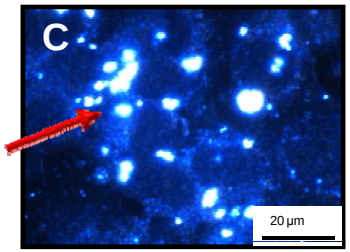
Uptake of TiO_2 Using CytoViva and TEM Imaging



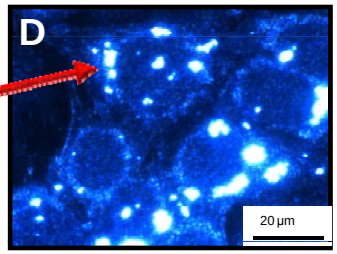
Untreated
CytoViva, 96X



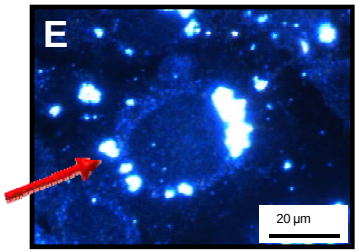
40 nm Amorphous TiO_2
CytoViva, 96X



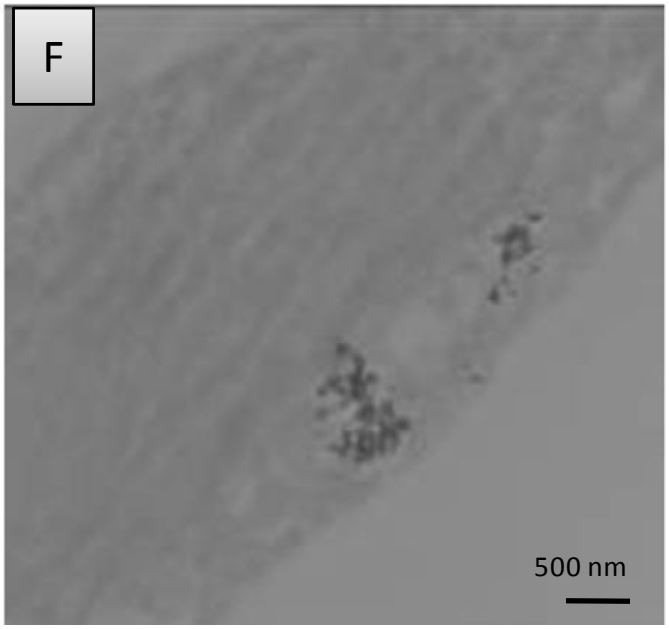
39 nm 40% Anatase TiO_2
CytoViva, 96X

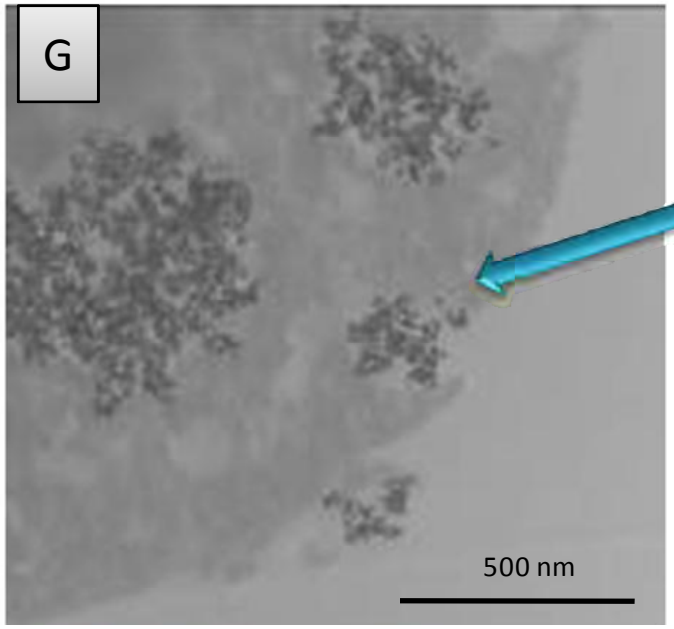


39 nm 61% Anatase TiO_2
CytoViva, 96X



100% Anatase TiO_2
CytoViva, 96X





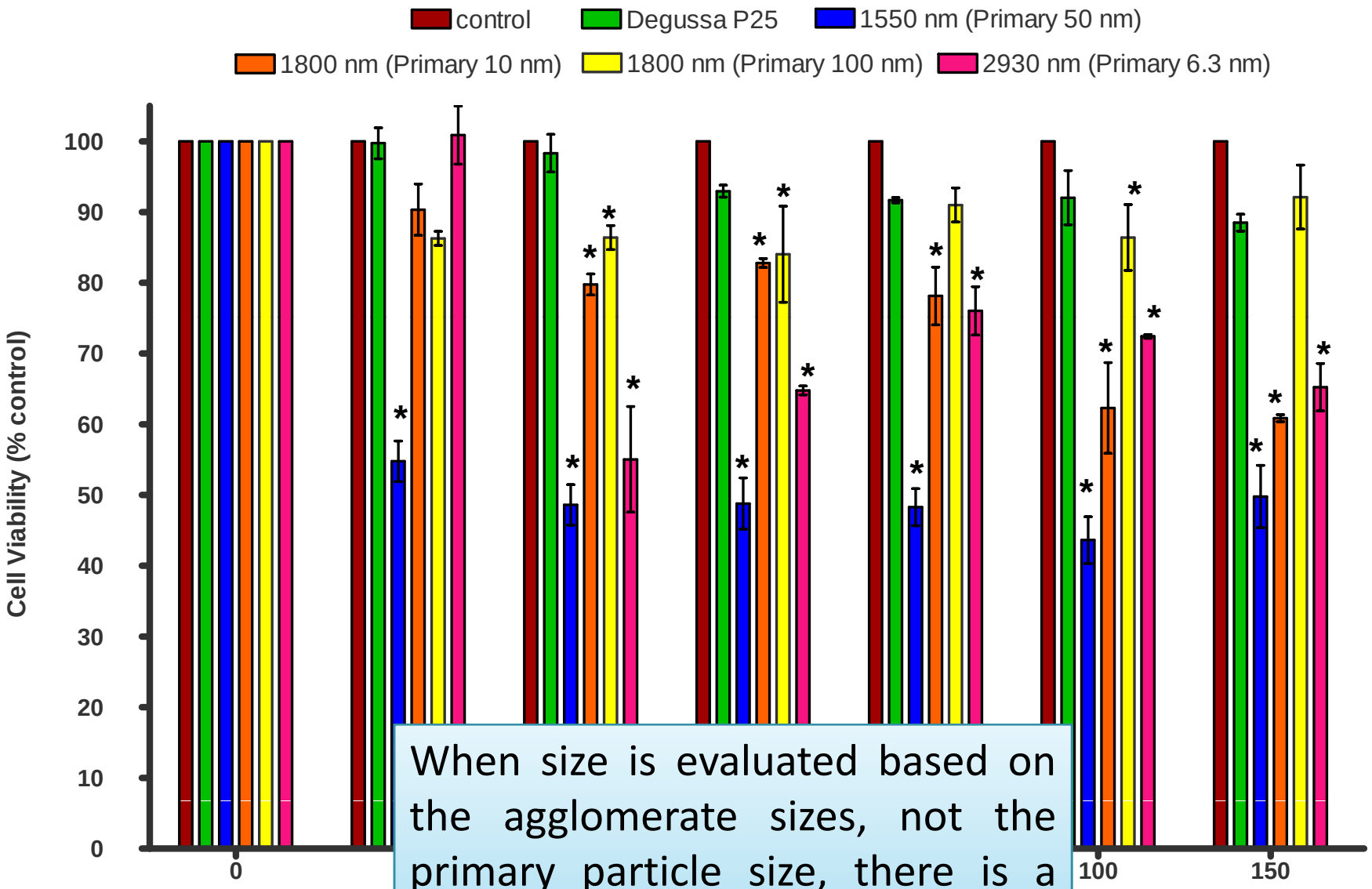
TEM of the 61% anatase

Cell appears to be "eating" the NP, indicates endocytosis as most likely mechanism of uptake.

Size vs. Crystal Structure in TiO_2 Nanotoxicity

	Particle	DLS		LDV	
		Average Diameter (nm)	PDI	Zeta Potential ζ (mV)	Electrophoretic Mobility U (μmcm/(Vs))
Crystallinity	***TiO_2 40 nm Amorphous***				
	DI H_2O	1300	0.282	-21.2	-1.66
	DMEM/F-12 Media	2040	0.349	* * *	* * *
	TiO_2 39 nm, 39% R, 61% A				
	DI H_2O	796	0.654	-23.3	-1.83
	DMEM/F-12 Media	2510	0.408	* * *	* * *
	TiO_2 39 nm, 60% R, 40% A				
	DI H_2O	519	0.661	-20.1	-1.58
	DMEM/F-12 Media	2030	0.743	* * *	* * *
	TiO_2 50 nm 100% A				
	DI H_2O	749	0.435	-13.7	-1.07
	DMEM/F-12 Media	1550	0.861	* * *	* * *
	TiO_2 51 nm, 100% R				
	DI H_2O	582	0.604	-21.8	-1.71
	DMEM/F-12 Media	1110	0.647	* * *	* * *
Size	***TiO_2 6.3 nm***				
	DI H2O	476	0.552	-29.0	-2.27
	DMEM/F-12 Media	2930	1	* * *	* * *
	TiO_2 10 nm				
	DI H2O	216	0.439	-2.79	1.63
	DMEM/F-12 Media	1800	0.402	* * *	* * *
	TiO_2 50 nm				
	DI H2O	749	0.435	-13.7	-1.07
	DMEM/F-12 Media	1550	0.861	* * *	* * *
	TiO_2 100 nm				
	DI H2O	1000	0.301	-21.3	-1.67
	DMEM/F-12 Media	1800	0.402	* * *	* * *
Cont'd	***TiO_2 Degussa***				
	DI H2O	542	0.499	19.4	1.52
	DMEM/F-12 Media	3500	0.303	* * *	* * *
	TiO_2 Ruthenium				
	DI H2O	663	0.689	-17.9	-1.41
	DMEM/F-12 Media	5870	1	* * *	* * *

Size vs. Crystal Structure in TiO_2 Nanotoxicity



When size is evaluated based on the agglomerate sizes, not the primary particle size, there is a size dependent effect observed.

Size vs. Crystal Structure in TiO_2 Nanotoxicity

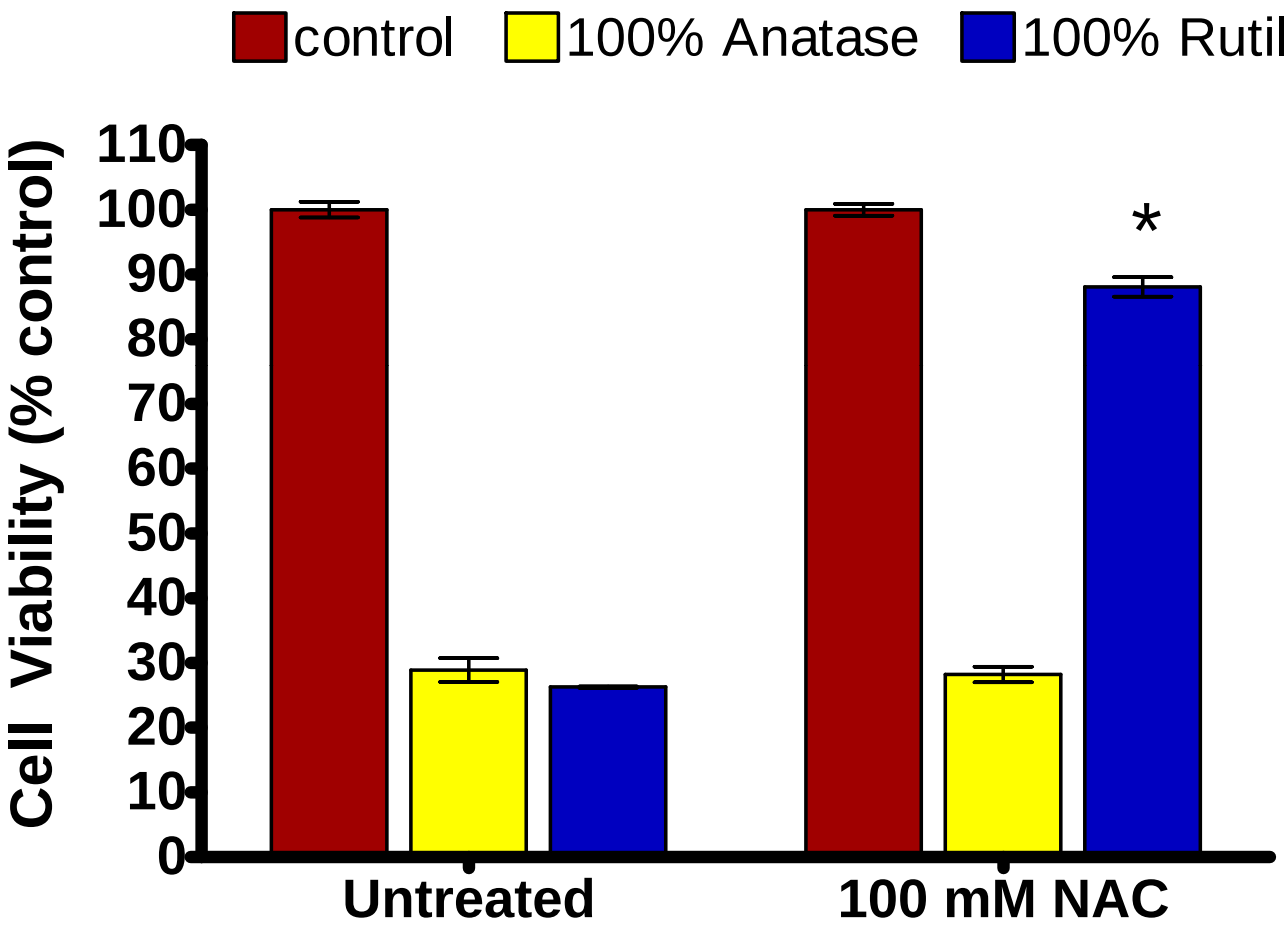
Summary of Cellular Effects Data

	Particle	Nanoparticle Characterization				Cellular Response to Nanoparticles			Mode of Cell Death
		TEM Average Diameter (nm)	DLS Hydrodynamic Diameter (nm)	LDV Zeta Potential (mV)	Acellular ROS Fold Increase over Control at 100μg/mL	MTS % to Control at 100μg/mL	LDH % to Control at 100μg/mL	ROS Fold Increase over Control at 100μg/mL	
Crystallinity	*TiO_2 40nm Amorphous*	40 ± 16	2040	-21.2	1.04	40.98 ± 7.97	3.71 ± 0.34	2.83	Apoptosis
	TiO_2 39nm, 39%R, 61%A	39 ± 10	2510	-23.3	1.42	32.12 ± 2.36	2.12 ± 0.12	1.74	Apoptosis
	Ti							3.29	Apoptosis
								1.32	Necrosis
								5.13	Apoptosis
Size								0.84	Necrosis
								0.95	Necrosis
								1.32	Necrosis
	TiO_2 100nm	100 ± 23	1800	-21.3	1.00	75.48 ± 0.13	124.50 ± 30.16	0.52	Necrosis
Control	*TiO_2 Degussa P25*	30	3500	19.4	1.73	91.92 ± 5.43	0.39 ± 0.22	1.75	Not toxic
	TiO_2 Ruthenium	40 ± 14	5870	-17.9	* * *	* * *	* * *	* * *	***
	Cellular Impact Level				**Low**	**Moderate**		**High**	

If the apoptosis correlates to the formation of ROS can antioxidants control this effect?

Crystal structure appears to be mediating the mechanism of cell death

Size vs. Crystal Structure in TiO_2 Nanotoxicity



Conclusion: The rutile TiO_2 ROS initiated apoptosis can be controlled for by treatment with antioxidants, thus making the anatase structure more toxic than the rutile.

Summary and Conclusions

The TiO2 nanoparticles are being taken up by the keratinocytes, most likely, through endocytosis.

The TiO_2 nanoparticles agglomerate when dispersed in exposure media.

When describing size dependent toxicity, agglomerate size and primary particle size must be taken into account

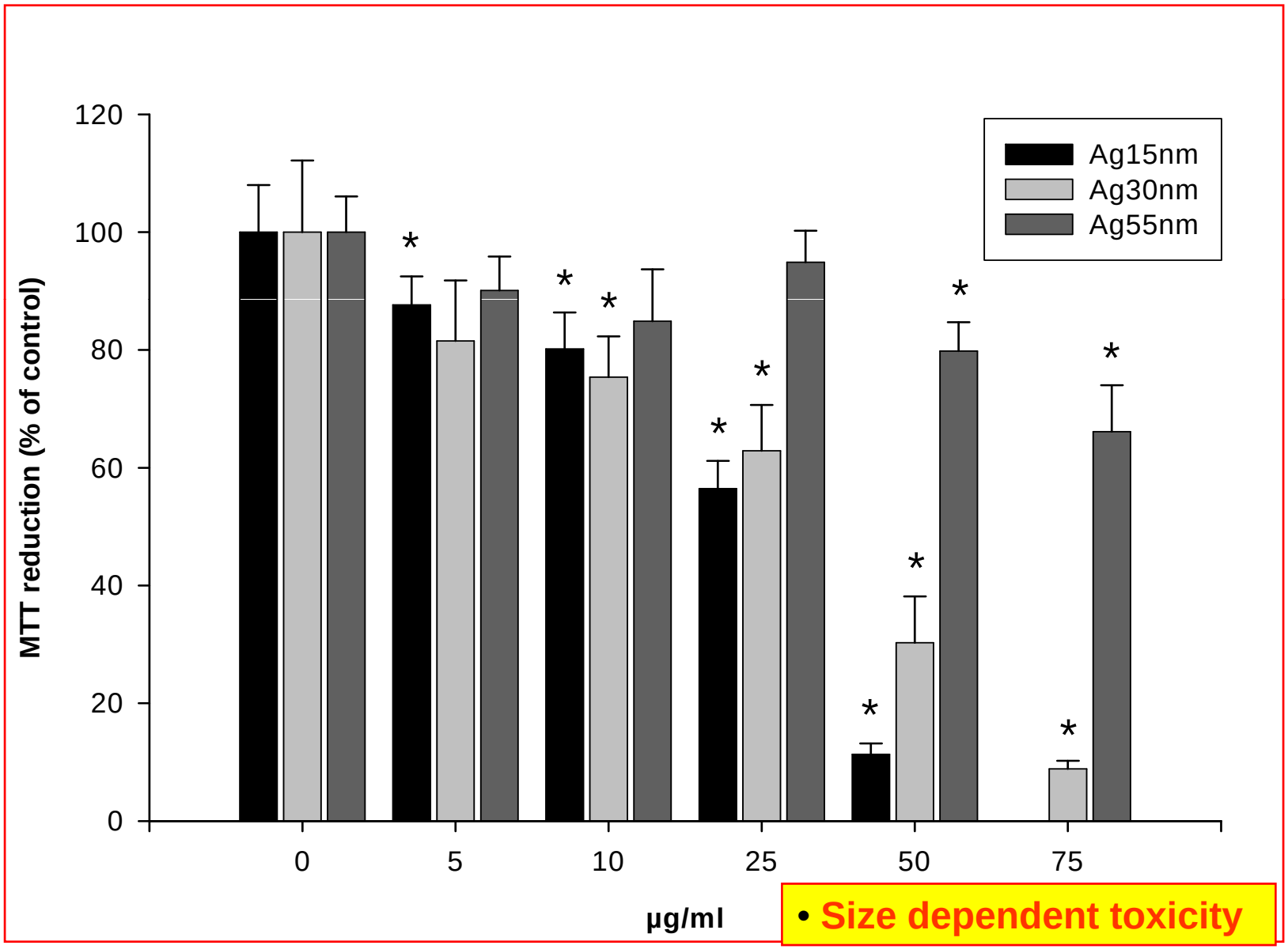
Crystal structure appears to be determining the type of cell death

High LDH leakage was associated with the anatase but not the rutile nanoparticles (indicates necrosis).

High levels of ROS production was associated with the rutile but not the anatase nanoparticles (indicates apoptosis).

Antioxidants can control the cell death induced by the rutile nanoparticles

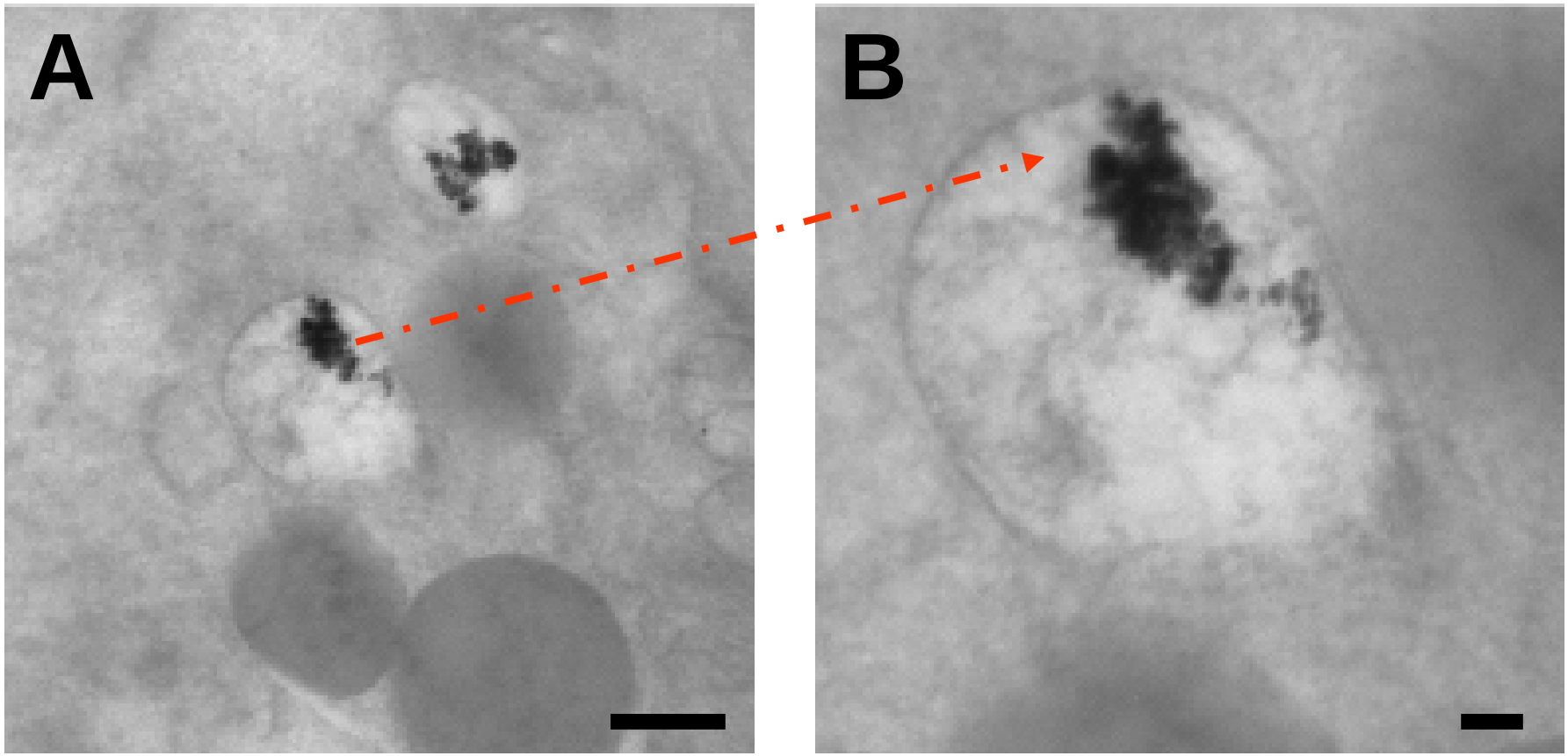
Toxicity of Silver NP Depends on Particle Size



- **Size dependent toxicity**

Biological Interaction of Nanomaterials

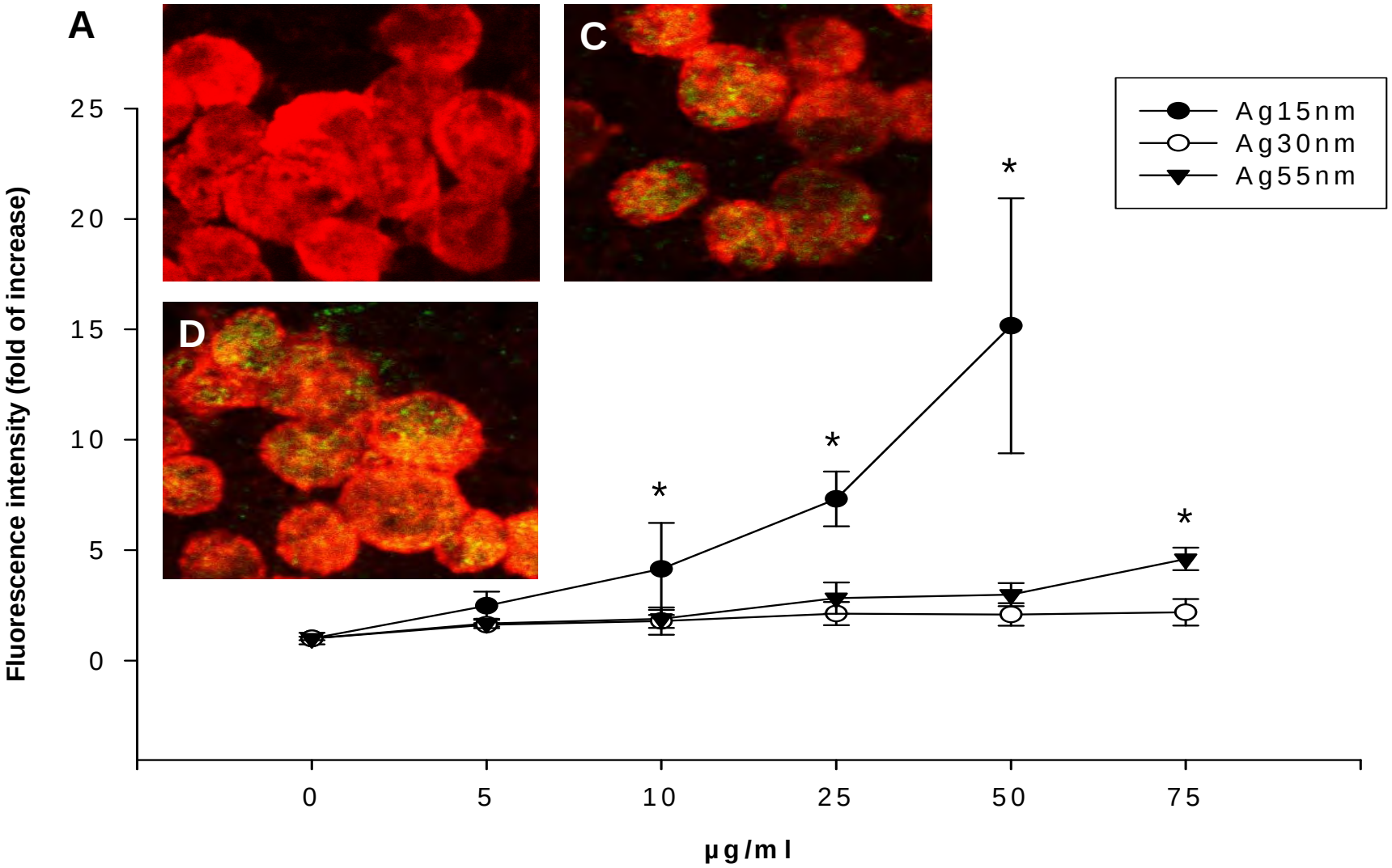
Silver Nanoparticles (Ag-NP)



Internalization and localization of Ag nanoparticles to intracellular vacuoles demonstrated by TEM:

Can Silver Nanoparticles be Useful as Potential Biological Labels?

Silver-NP induced ROS Generation Based on Size



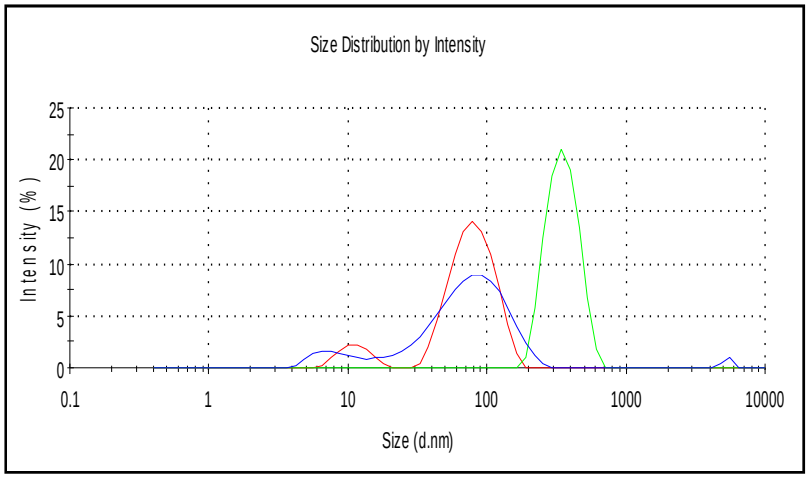
- **Size dependent induction of oxidative stress**

Nanoparticle Coating vs. Toxicity

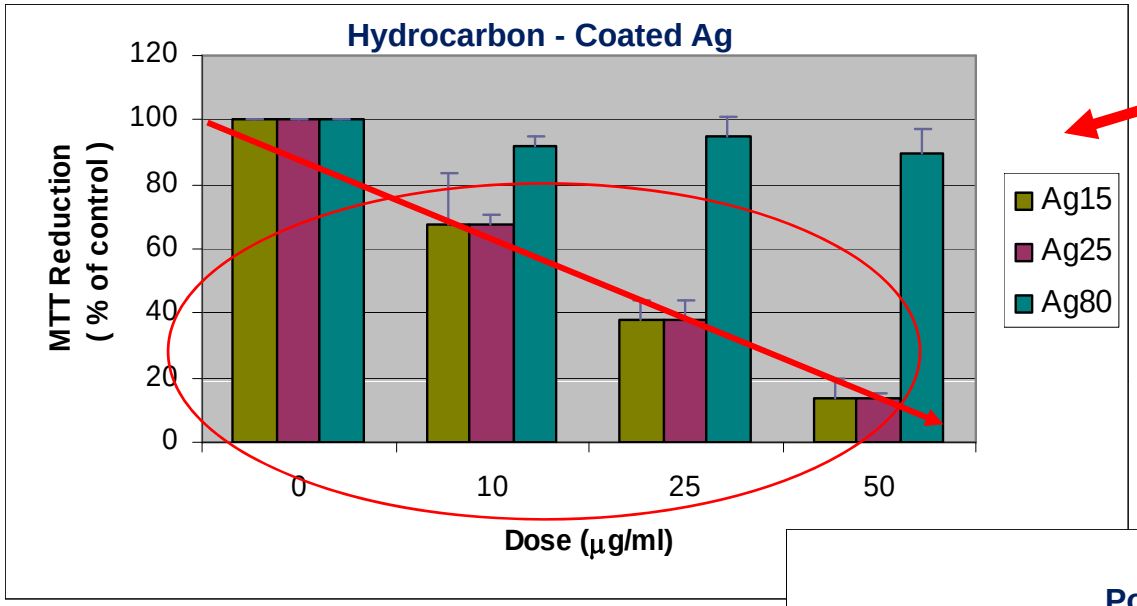
PS coated Silver Nanoparticles: Size Determination by DLS

DLS

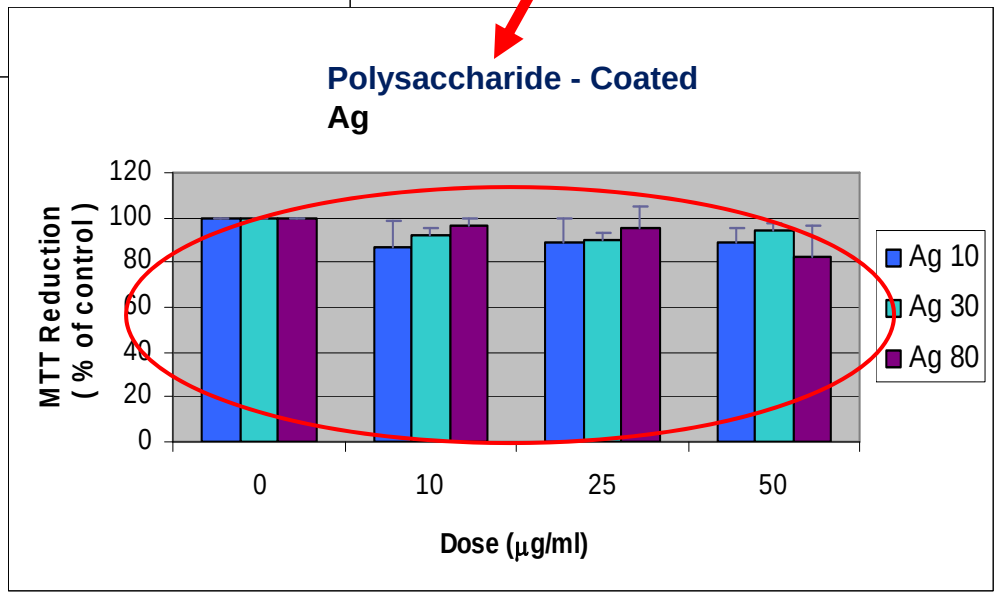
Particle	Average diameter (nm)
PS-Ag 10 nm	
DI H_2O	72.8
RPMI-1640 media	413
RPMI-1640 media wt/ 20% serum	49.4
PS-Ag 25–30 nm	
DI H_2O	128
RPMI-1640 media	261
RPMI-1640 media wt/ 20% serum	118
PS-Ag 80 nm	
DI H_2O	250
RPMI-1640 media	743
RPMI-1640 media wt/ 20% serum	1230



Surface Coating Protect from Silver-NP Toxicity

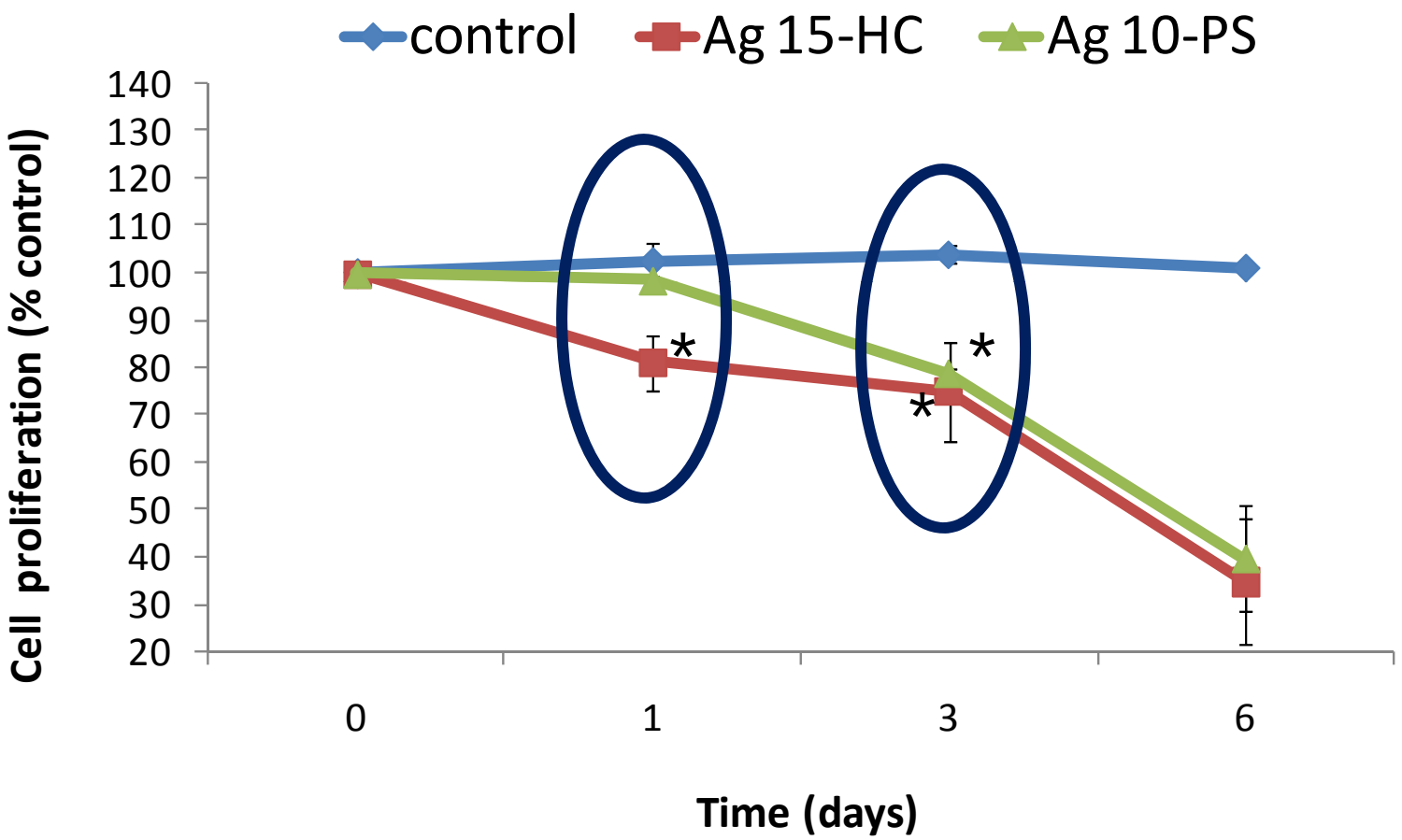


Different coatings decreased toxicity of Ag nanoparticles



Cell viability was significantly higher when dosed with PS-Ag, even at 50 µg/mL, when compared to HC-Ag samples.

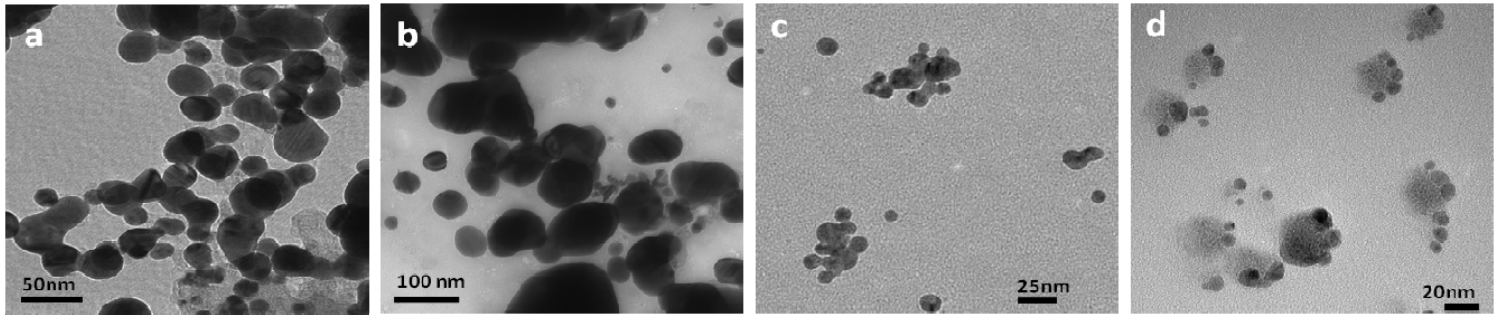
Is PS Coating Stable?



Mostly likely cause---degradation of NP coating

Braydich-Stolle et al., submitted

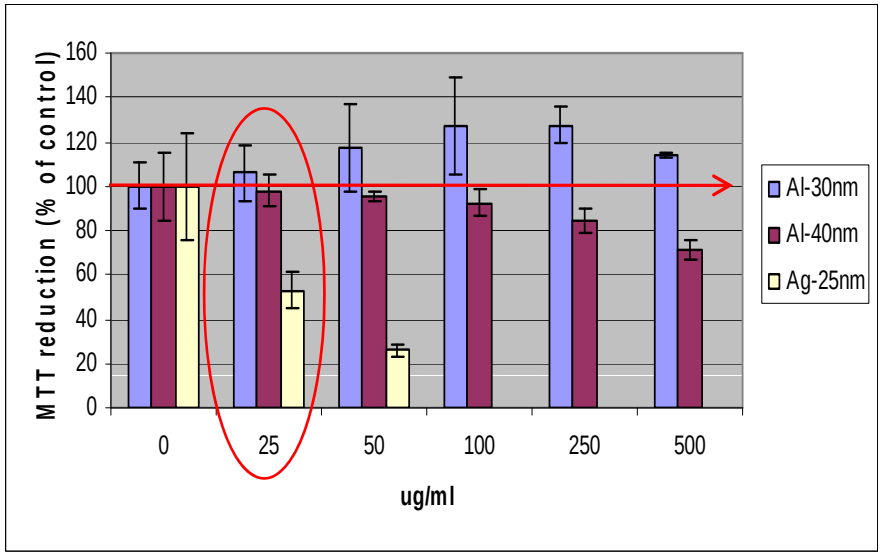
Characterization of Coated Silver-NP

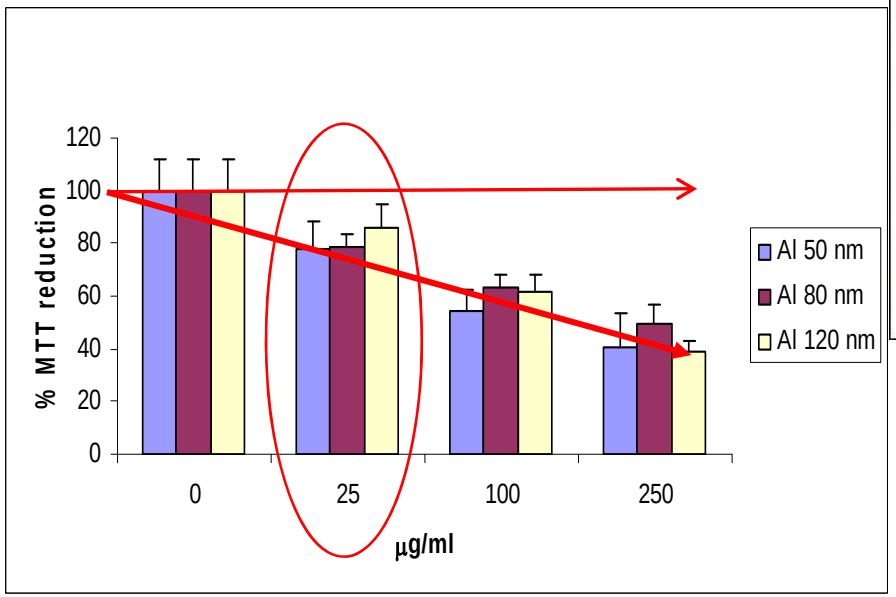


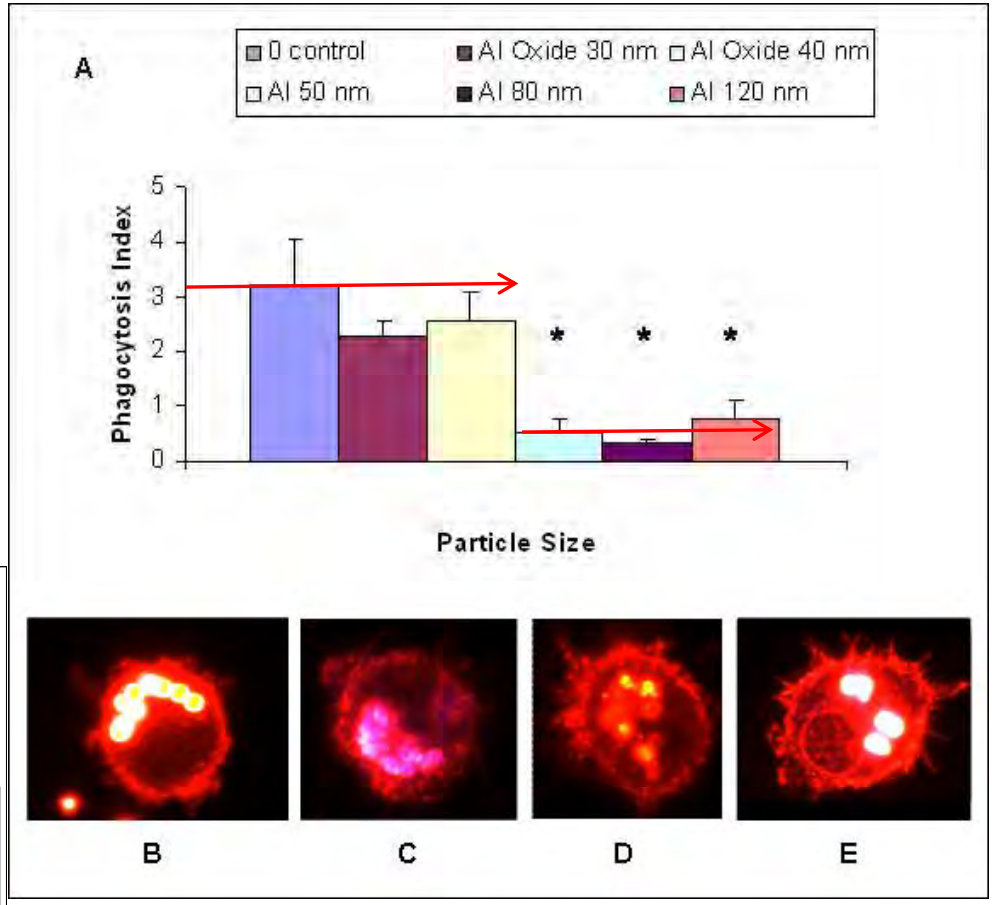
e Sample	Mean Primary Particle Diameter ± SD (nm) (TEM)			Z-Average Particle Diameter (nm) (DLS)	
	Pre-Exposure	Intracellular	Post-Exposure	Pre-Exposure	Post-Exposure
Ag 25-HC	32.5 ± 12.4	20.1 ± 10.3	34.4 ± 22.8	208*	155
Ag 10-PS	12.0 ± 3.9	34.6 ± 7.8	6.9 ± 2.2	72.8*	298

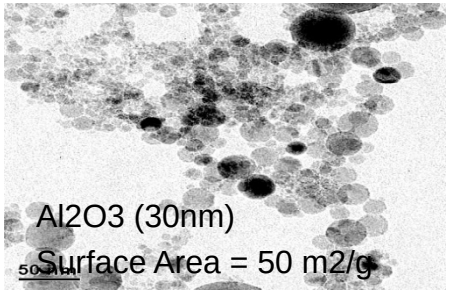
f	Sample	C			O	Na	N	Ag	Al	Cl	S
		C=O	C-O	C-H, C							
Pre-Exposure	*Ag 25-HC*										
	-1	4.9	7.6	28	33.5	- - -	- - -	22.4	3.7	- - -	- - -
	Ag 10-PS										
	-1	1.2	3.8	62.6	20	9	0.5	0.2	- - -	- - -	2.7
	-2	1.2	4.3	61.8	20.2	8.8	0.7	<0.1	- - -	- - -	2.9
Post-Exposure	*Ag 25-HC*										
	-1	3.7	11.3	30	20.9	- - -	- - -	30.4	- - -	3.8	- - -
	-2	4.9	7.2	23.6	33.5	- - -	- - -	27.4	- - -	3.5	- - -
	Ag 10-PS										
	-1	4.8	12.7	8.4	44.4	- - -	- - -	28.2	- - -	1.4	- - -
	-2	4.6	16.2	7.2	42.7	- - -	- - -	27.5	- - -	1.9	- - -

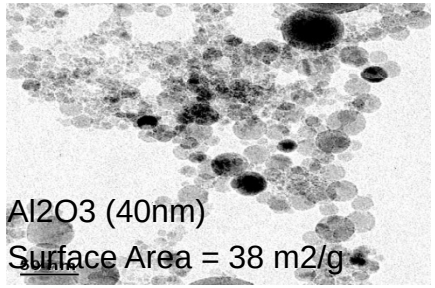
Toxicity of Al-NP Depends on Surface Coating







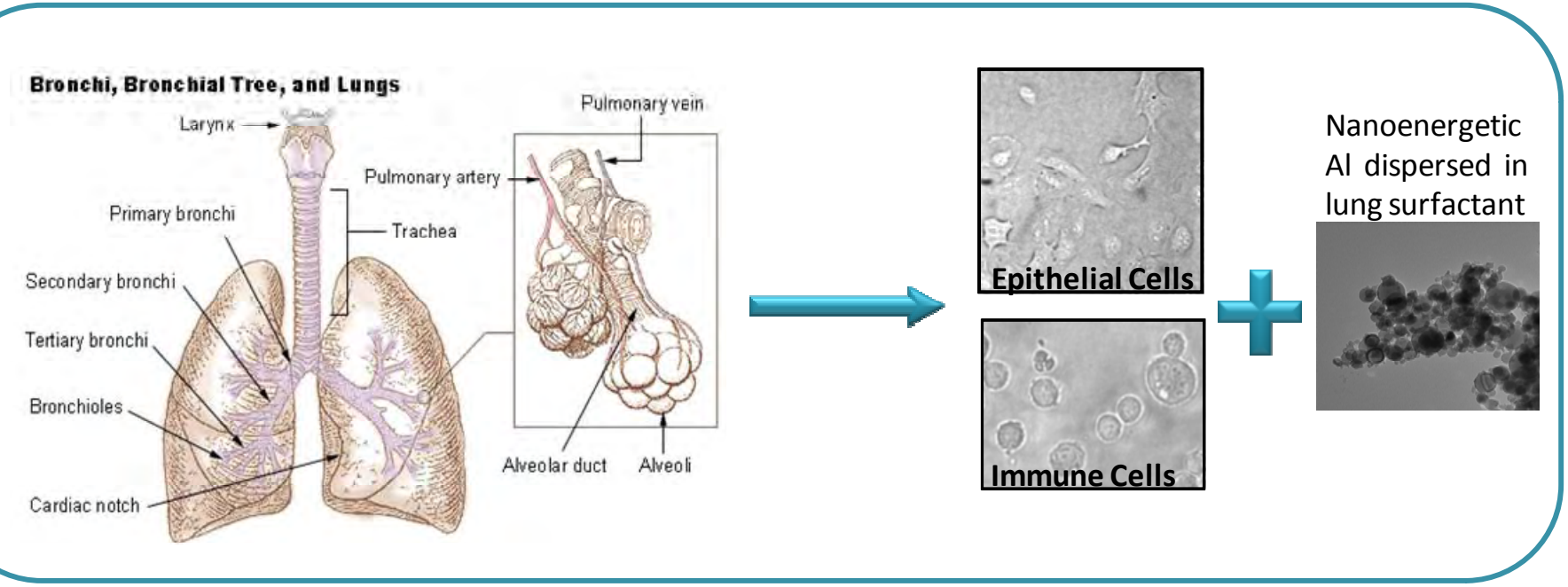




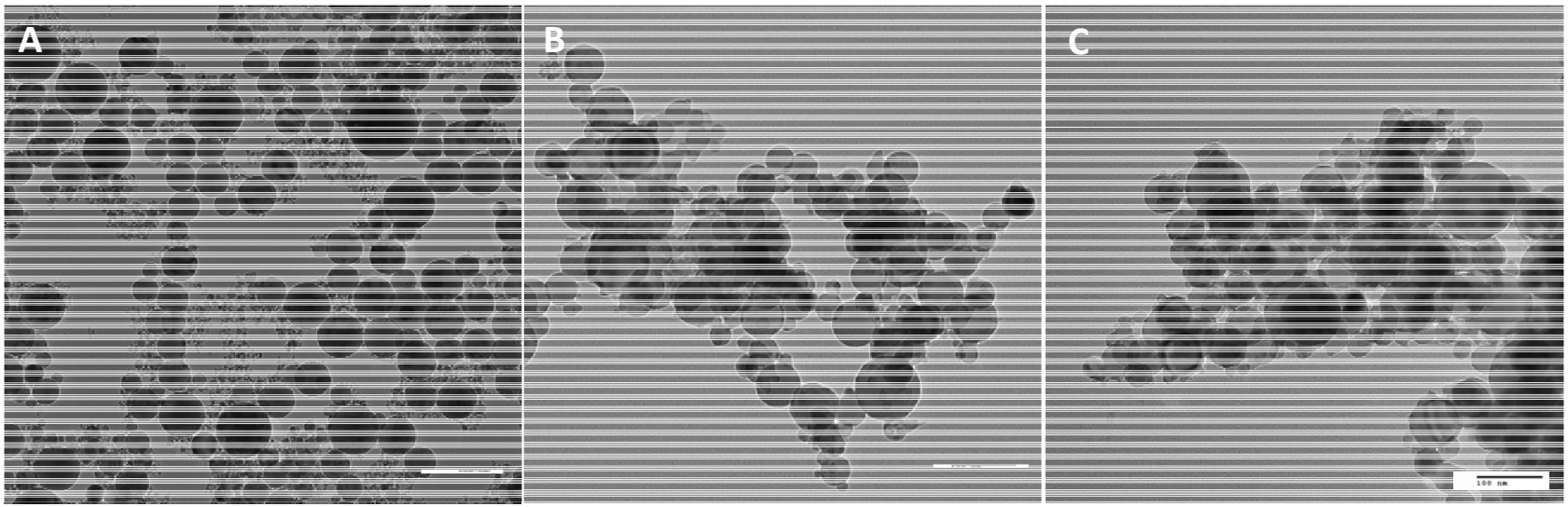
Evalutation of Nanoenergetic Aluminum

Inhalation the most likely exposure scenario.

Development of co-culture model to assess cell viability, phagocytic activity, activation of immune response, and secretion of inflammatory responses.



Characterization of Nanoenergetic Aluminum



D

Sample	Primary Nanoparticle Size TEM (nm)	DLS Z-Ave (d.nm) ± PdI			
		Dispersion in Water		Dispersion in Artificial Lung Surfactant	
		Exposure Media	Growth Media	Exposure Media	Growth Media
Al_2O_3-40nm	48.08 + 21.01	859 ± 0.25	309 ± 0.359	878 ± 0.495	486 ± 0.603
Al-50nm	32.71 + 28.28	698 ± 0.598	839 ± 0.661	805 ± 0.497	948 ± 0.618
Al-OA-50nm	51.09 + 22.48	5700 ± 1.0	138 ± 0.179	2430 ± 1.0	195 ± 0.307

Establishment of Co-culture

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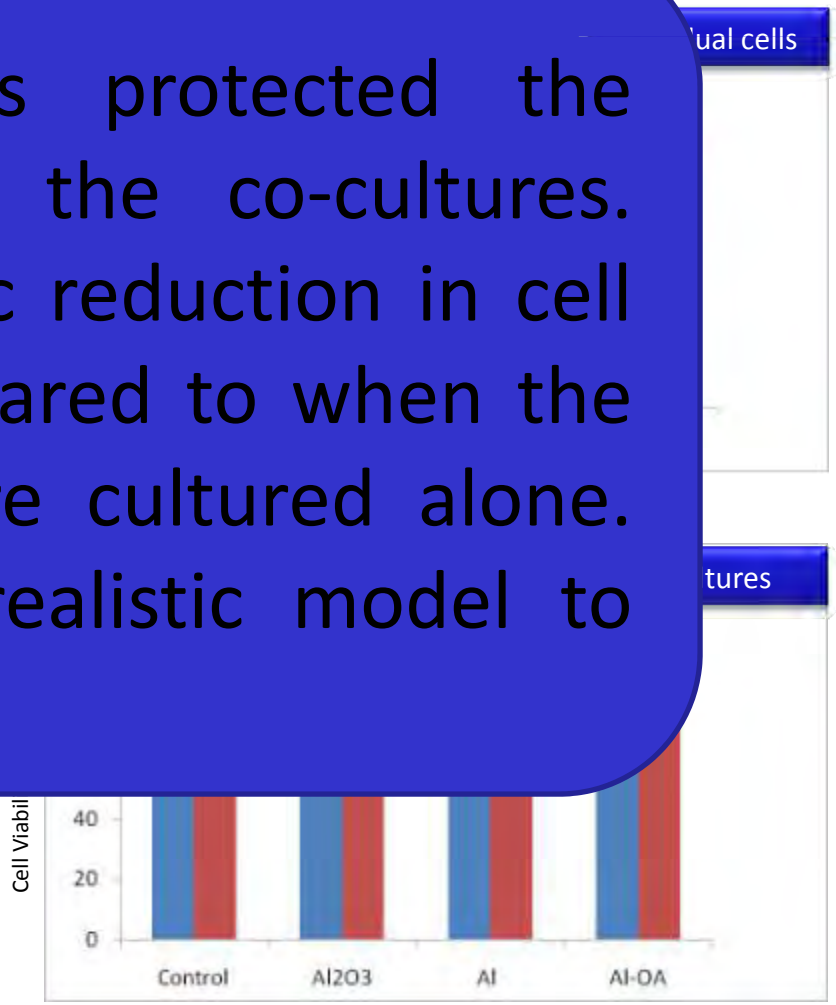
3:1 ratio of A549:U937 cells[1]

The nanoparticles were dispersed in an artificial lung surfactant[2]

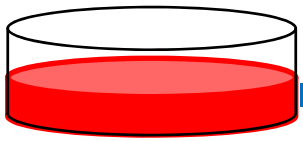
[1] Wang et al. (2002) Toxicology 173: 211-219.
[2] Ansoborlo et al. (1999) Health Physics 77: 638-645

Cell were treated with 25 µg/ml of nanoparticles

The immune cells protected the epithelial cells in the co-cultures. There was a drastic reduction in cell death when compared to when the epithelial cells were cultured alone. Provides a more realistic model to assess exposure.

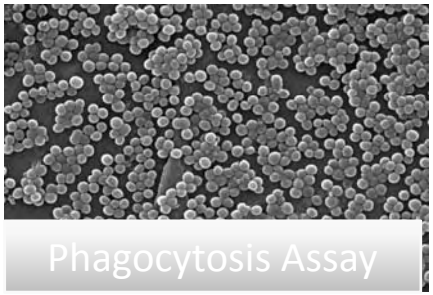


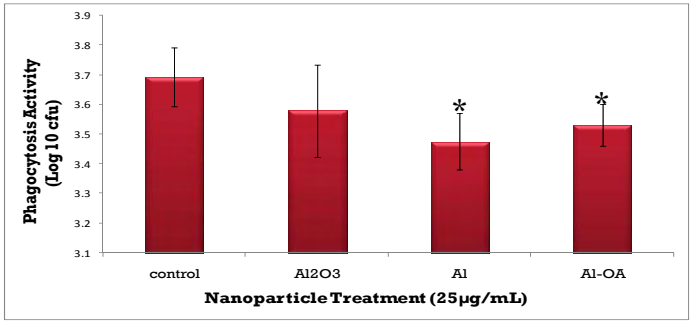
Evaluation of Macrophage Function



24 h Al NP Exposure

25 µg/ml



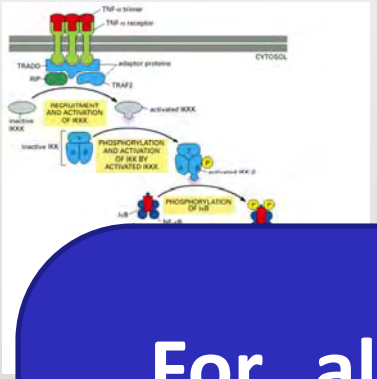


Al reduced the macrophages ability to phagocytose bacteria.

Once phagocytosis occurs the NFκB pathway is activated to produce inflammatory cytokines.

Will the reduction in phagocytic function impact the NFκB pathway and cytokine secretion???

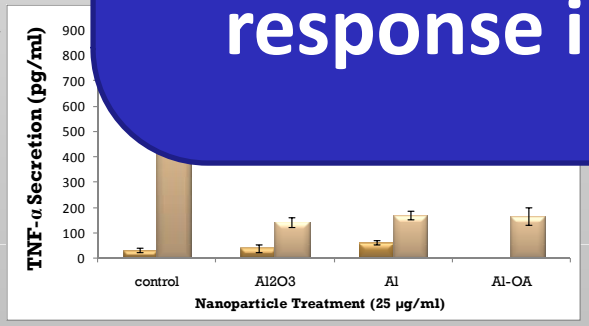
Secretion of Inflammatory Cytokines

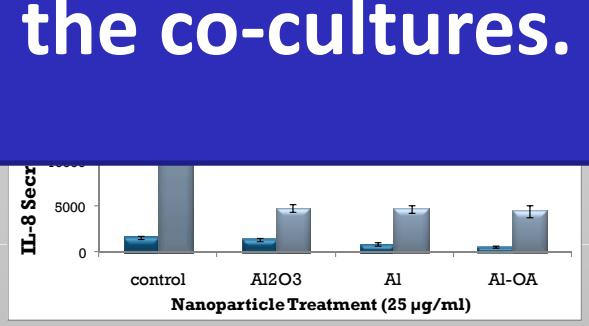


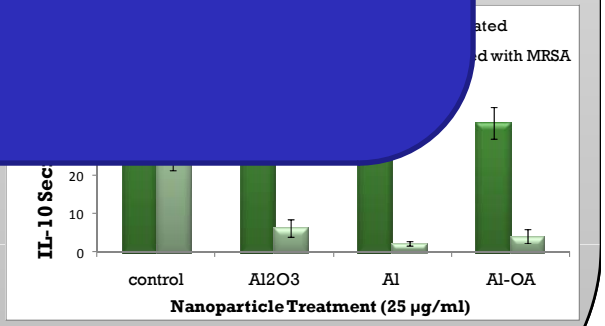
Secretion of IL-6, IL-1β, TNFα, IL-8, and IL-10 was evaluated.

For all 5 cytokines assessed, the Al nanoparticles caused a massive down-regulation in the secretion of cytokines. The Al altered the immune response in the co-cultures.

C







Summary

Established a co-culture system to simulate the alveolar microenvironment

The Al nanomaterials show more localization in the immune cells in comparison to the epithelial cells.

When a respiratory pathogen is introduced into these co-cultures there is a difference in how these cells respond when the Al nanoparticles are present.

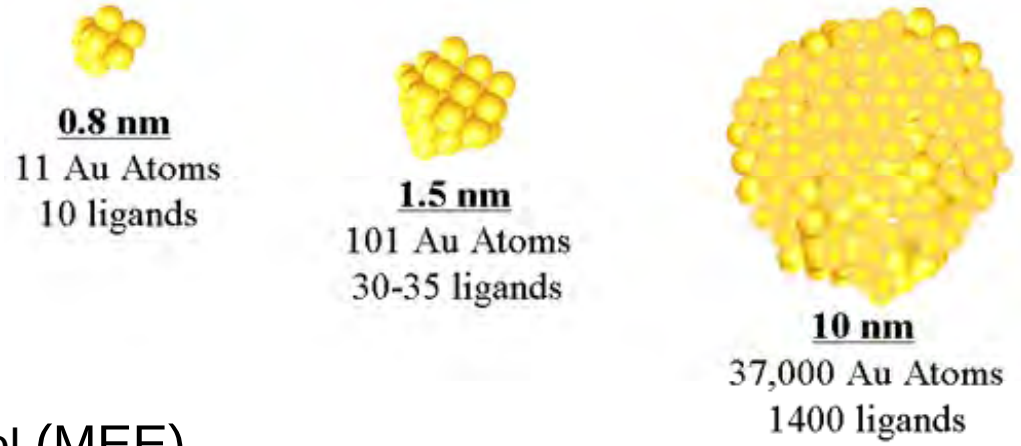
Cytokine secretion is comparable to control levels in the Al NP treatments even when infected with MRSA.

Despite the low toxicity, the presence of the Al NPs interferes with the cells natural ability to respond to pathogens

Surface Charge vs. Toxicity

Gold Nanoparticles: Size and Surface Functionalization

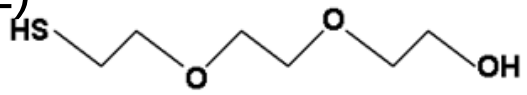
Courtesy of Bettye L.S. Maddux, Ph.D.
University of Oregon, Materials Science Institute



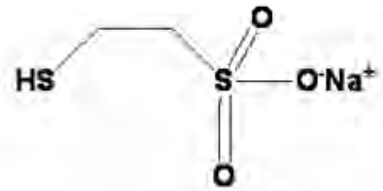
Surface Functionalization

Neutral: 2-(2-mercaptoethoxy)ethanol (MEE)
0.8 and 1.5 nm AuNPs

Neutral: 2,2,2-[mercaptoethoxy(ethoxy)]ethanol (MEEE)
0.8, 1.5 and 10 nm AuNPs

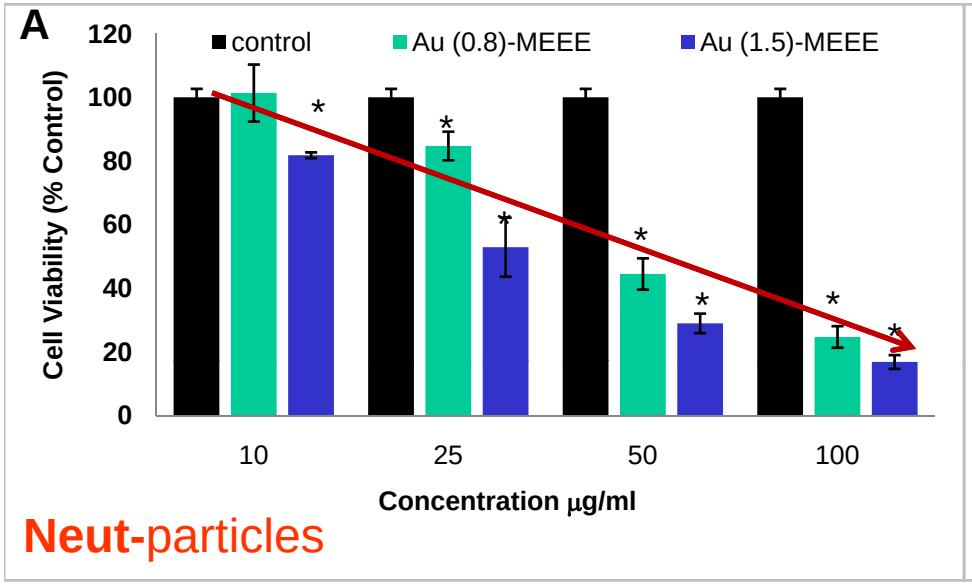


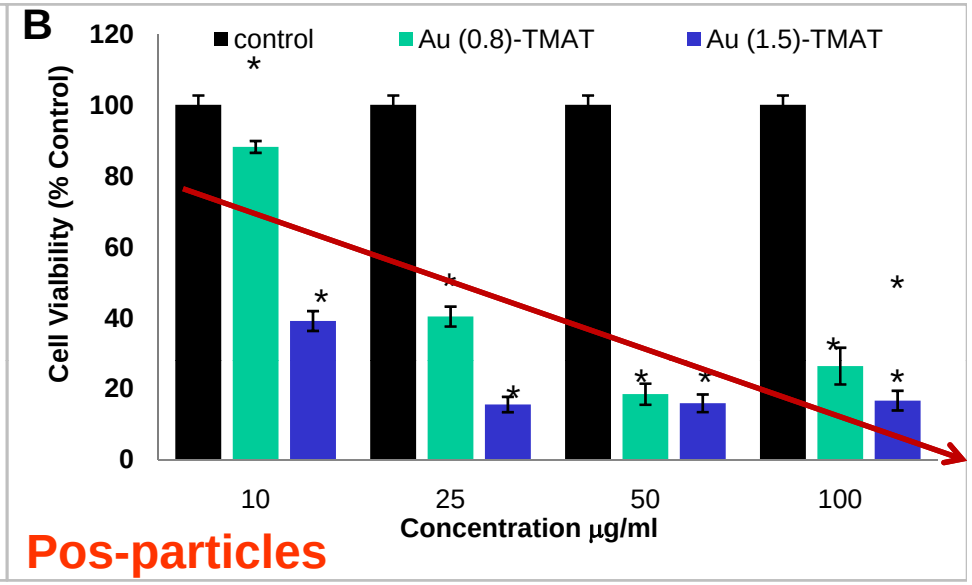
Anionic: 2-mercaptoethanesulfonate (MES)
0.8 and 1.5 nm AuNPs

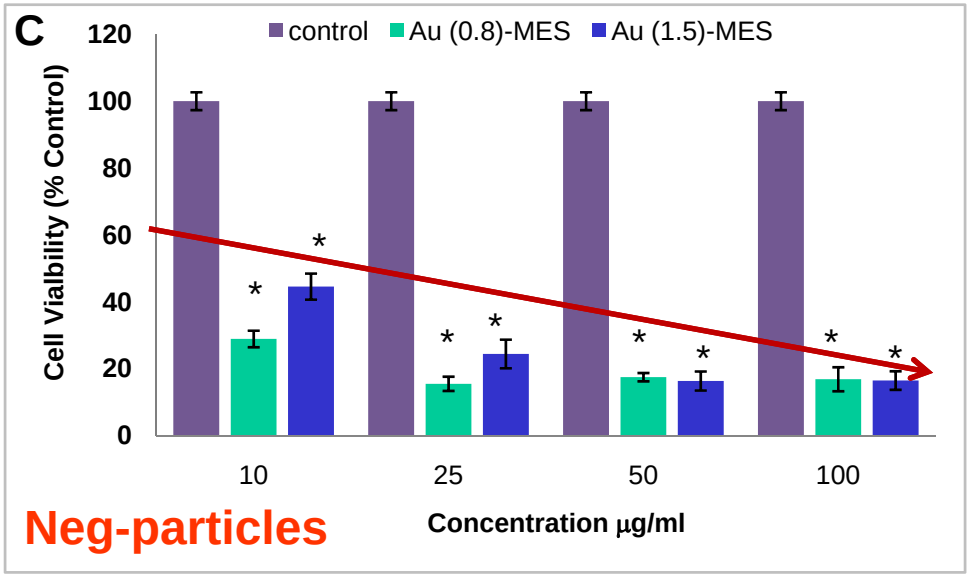


Cationic: N,N,N-trimethylammoniumethanethiol (TMAT)
0.8, 1.5 and 10 nm AuNPs

Toxicity of Au-NP Depends on Surface Charge







Summary of Characterization!!!

- **Characterization of nanomaterials before and after exposure**
- **Supply of well characterized nanomaterials**
- **Toxicity dependent on size, coating, charg & Shape**
  - **Size: Ag (15, 25, 55 nm) = Size dependent toxicity**
  - **Size: Ag (15 nm) induces oxidative stress**
  - **Agglomeration: Al (50,80,120 nm) decreased phagocytosis but not size dependent**
  - **Coating: Ag-PS (10, 25-30 nm) = No toxicity (> 100ug/ml)**
  - **Coating: Al_2O_3 (30, 40 nm) = Not toxic**
  - **Charge & Shape: Gold Particles**

Characterization of nanomaterials (understanding surface Properties) is key to establishing safety of nanotechnology

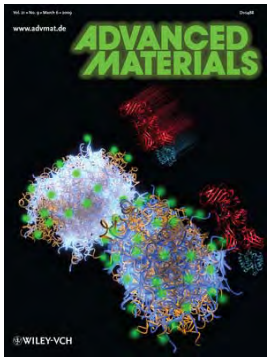
What's the Message?

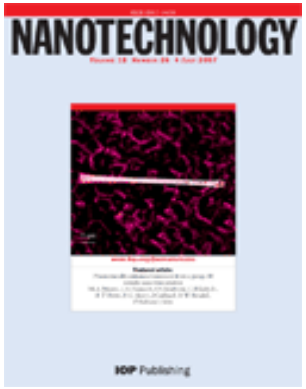
- Size matters, but not always
- Physicochemical character matters
  - Crystallinity
  - Chemical reactivity
  - Shape, charge
  - Coating
- Contaminants must be considered
- Agglomeration vs. dispersion- Critical point
- Charge matters (affects reactivity and dispersion)

Good Nanotoxicology Requires

Good Characterization

Scientific Impact





Schrand A, Braydich-Stolle L, Schlager LL, Hussain SM. Can silver nanoparticles be useful as potential biological labels? Nanotechnology 19 (June 11 2008) 235104

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Adv Mater. 2009 (2): 1-11.

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Thank you

Questions??

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